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An investigation of motor disabilities in people with multiple sclerosis using advanced magnetic resonance imaging

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An investigation of motor disabilities in people with multiple sclerosis using advanced magnetic resonance imaging

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door

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CHAPTER 1

General introduction

Epidemiology and disease course of multiple sclerosis

Multiple sclerosis (MS) is a progressive autoimmune disorder of the central nervous system (CNS), and is the most common cause of neurological disability in young and middle-aged adults, ranging between 20 and 40 years old. The pathology of MS is caused by an unknown aetiology and is characterized by inflammation, demyelination and axonal injury and loss within the brain and spinal cord. The disease affects more women than men with ratios as high as 3:1, and has a huge impact on participation and quality of life. More than 2.5 million people world-wide suffer from MS, which makes it a major public health concern.[1] While MS can occur across the globe, prevalence varies with ethnicity and location plays a role. Particularly people in the northern parts of Europe, North America and Australia appear to be highly susceptible to develop MS.[2,3]

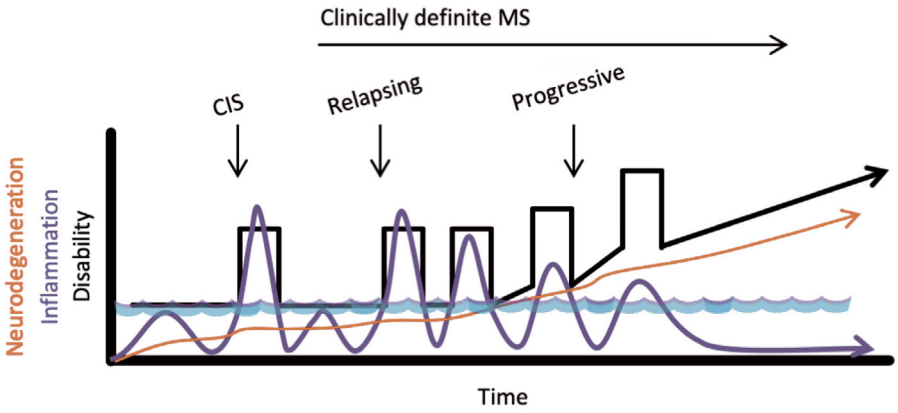


Figure 1. Example of multiple sclerosis disease course. A clinical presentation suggestive of multiple sclerosis is called a clinically isolated syndrome (CIS). Most people get diagnosed with relapsing-remitting multiple sclerosis, which is predominantly driven by relapses and characterized by high clinical and subclinical inflammatory disease activity. The relapsing-remitting disease course can transit into a secondary progressive stage, characterized by increase in neurodegeneration and worsening of disabilities.

Most people with MS (pwMS) have a biphasic disease course, starting with periods where neurological symptoms flare up (i.e. a relapse) and then mostly disappear (i.e. a remission), so called relapsing-remitting MS (RRMS) (Figure 1). Remission following an acute relapse often occurs within weeks to months and most pwMS experience complete recovery. However, recovery is incomplete in a minority of pwMS and becomes less over time leading to permanent disability. In approximately 65% of RRMS patients the disease evolves into a secondary more progressive stage characterized by irreversible, continuous neurological decline with minimal relapses and remissions (SPMS).[1] Conversion to SPMS happens after roughly 10 years in 50% of pwMS,[4] with older age and motor symptoms at RRMS onset associated with higher risk of conversion.[5] In a minority of pwMS, approximately 15%, MS is characterized by slow unremitting progressive deterioration from onset and irreversible disability, known as primary progressive MS (PPMS) (Figure 1).[1,4] Despite the distinction between relapsing-remitting and progressive stages, the clinical course of MS is highly variable and largely unpredictable.

Pathology of multiple sclerosis

MS is typically characterized by damage to the myelin in the white matter (WM) and grey matter (GM), also known as "lesions" or "scars". Myelin is a fatty sheath around axons, also known as nerve fibers, and is created by oligodendrocytes. These cells wrap myeline sheaths around several axons with gaps in between, known as the nodes of Ranvier, which improves signal conduction in the brain. MS not only alters axonal wiring in brain and spinal cord initially by demyelination of axons, but also involves axonal injury and loss. Subsequent to axonal injury, Wallerian (distal) and retrograde (proximal) degeneration can occur, eventually leading to neuronal death in cortical[6] and deep GM structures[7] and disconnections within and between brain networks. Whereas both WM and GM pathology can involve focal demyelinating lesions as well as neurodegeneration, GM differs from WM pathology in that immune cell infiltration is limited and absence of blood-brain barrier breakdown,[8,9] two typical pathological characteristics of MS.

Over the last decades extensive and comprehensive research has been done studying the aetiology of MS, but unfortunately the cause remains unknown. Suggested potential risk factors include environmental elements such as vitamin D deficiency, Epstein-Barr viral infection, smoking[2] and obesity in early life.[10] Even though MS is not considered as an inherited disease, recent genome wide association studies have discovered a possible genetic component. More than 200 risk loci have been implicated to contribute to MS pathogenesis.[11] While presumably the interplay between environmental and genetic factors play a role in the development of MS, most research evidently points towards dysregulation of the immune system.

Two major immune system hypotheses have been proposed, the "outside-in" and "inside-out" theory. With the "outside-in" theory MS is seen as a classical autoimmune disease where a primary trigger within the immune system leads to a malfunctioning system and an immune response is directed against brain and spinal cord leading to inflammatory and demyelinated lesions.[1,12] However, one single specific event leading to pathology within the CNS has not been identified.[13] Alternatively, the inside-out theory suggests that neuroaxonal or oligodendrocyte disturbances in the brain could be initial triggers that lead to immune response and focal areas of inflammatory lesions as secondary response.[14,15]

MRI as a clinical tool for diagnosis and identifying multiple sclerosis pathology

Magnetic Resonance Imaging (MRI) is a powerful tool in human neuroscience and has contributed significantly to our understanding of MS. In clinic, conventional MRI plays an imperative role in diagnosis and monitoring disease progression. The focal inflammatory demyelinating lesions in the WM, the classical pathological hallmark of MS, can be visualized using multiple MR sequences including a fluid-attenuated inversion recovery (FLAIR) scan, T1-weighted and T2-weighted imaging (Figure 2). The diagnostic criteria involves dissemination of lesions in space and time on MRI, in addition to clinical observation and laboratory assessment of oligoclonal bands in the cerebrospinal fluid.[16,17] A T1-weighted

image with gadolinium enhancing contrast can be used to differentiate between active lesions (enhancing) and chronic lesions (non-enhancing) (Figure 2). Whereas enhancing lesions are an indication of ongoing inflammatory processes,[18] hypointense lesions are suggestive of axonal destruction and irreversible damage, also known as black holes.[19] The presence of both T1-weigthed enhancing and non-enhancing lesions suggests that onset of lesions occurred at different times, which fulfils the dissemination in time criteria. To meet the dissemination of lesions in space criteria, one or more lesions must be present in at least two areas considered typical for the disease (periventricular, juxtacortical, infratentorial and spinal cord) (Figure 2).

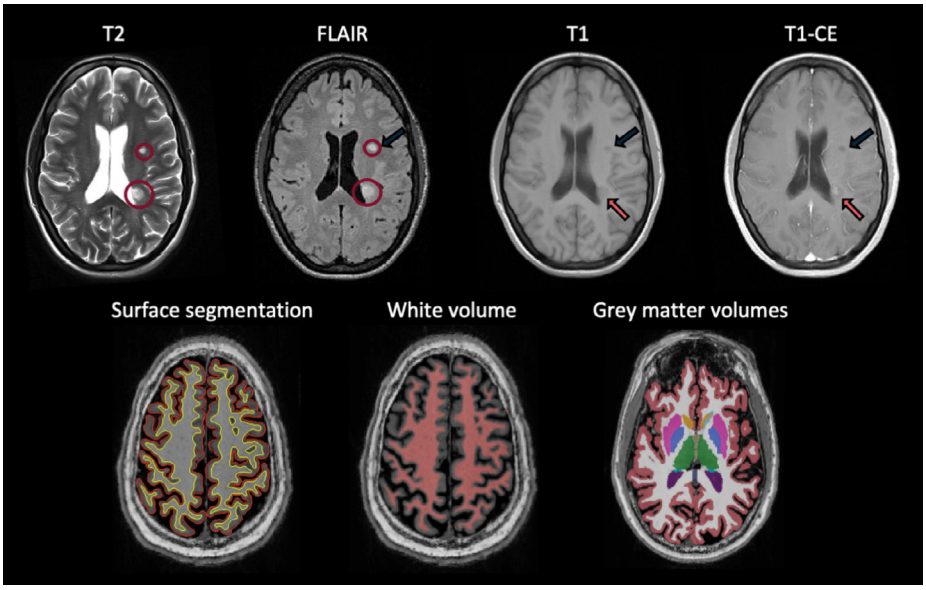


Figure 2. Current radiological practice and brain volumetrics. Four sequences that are often used in clinic for diagnosis and disease progression can include a T2-weigthed image, fluid-attenuated inversion recovery (FLAIR) scan and T1-weighted scan without and with contrast enhancing gadolinium (T1-CE). White matter lesions are clearly visible on a T2-weighted MRI and FLAIR scan as bright white spots in contrast to the darker surroundings suggestive of normal appearing WM (red circles). Contrast-enhanced imaging is a sensitive method to visualize blood brain barrier breakdown and enhancing active lesions, i.e. lesions in the inflammatory phase (pink arrows). A black hole appears hypointense on T1-weighted scan and hyperintense in the corresponding T2-weighted image (dark green arrows). Brain volumetrics can be measured using 3D T1-weighted image and segmentation methods such as FreeSurfer (https://surfer.nmr.mgh.harvard.edu/) and can include white matter, (sub) cortical grey matter and ventricular volume.

While visualizing focal WM lesions is essential in clinic for diagnosis, the relation between lesions and disability and cognitive dysfunction is relatively poor.[20,21] The dissociation between volume and number of lesions and clinical performance is known as the clinico-radiological paradox.[22] In other words, pwMS with different lesion loads can have similar symptomatologic presentations or the other way around. The paradox was a driver to not only focus on lesion pathology but to investigate other pathological processes including axonal injury/loss and GM pathology.

Apart from WM and GM inflammatory lesions, a very prominent and important pathological component of MS is neurodegeneration. A marker of neurodegeneration is brain atrophy, which can be measured using 3D-T1 weighted imaging (Figure 2). Brain atrophy can be quite extensive with a yearly rate of 0.5-1.35% and proceeds substantially faster compared to normal aging.[23,24] Whereas GM atrophy appears to predominate over WM atrophy and progressively accelerates after conversion to SPMS, the rate of change in WM atrophy remains constant over time,[25] suggesting partially independent pathological processes. A reduction in WM volume may at least partly reflect lesion volume reduction due to axonal degeneration.[13,26] The histopathological substrate of GM atrophy could be neuroaxonal cell loss due to either retrograde degeneration as consequence of WM axonal transection or due to pathological processes within GM.[26,27] The predominant pathological substrates of MRI-measured GM atrophy was suggested to involve neuronal and axonal pathology including reduced neuronal size, neuronal and axonal density.[28]

Whereas the number and volume of lesions explain variability in disability only to a small extend,[21,22] stronger relations have been observed with GM volume loss.[29,30] Brain atrophy relates to clinical impairments from onset[31] with progression of disability more closely correlated to GM atrophy than loss of WM volume.[32] Regional analyses have highlighted the importance of damage to specific structures in the progression of clinical impairments. Atrophy of the deep GM structures relates strongly to disability, particularly the thalamus plays an important role in MS.[33] Thalamic atrophy precedes clinical symptoms[34] and progresses rapidly throughout the disease with strong correlation to disability progression.[33] Besides brain volumetrics, spinal cord damage is commonly observed in MS but often overlooked. Spinal cord damage is not always readily apparent in early RRMS patients[35] and is greater in those in the progressive stage of MS.[36]

Symptomatology of multiple sclerosis

This disseminated nature of MS pathology leads to a wide range of symptomatologic presentations. The presenting signs, degree of disability and disease course are highly heterogeneous both within and between pwMS. An example of a common manifestation and presenting sign of MS is optic neuritis, characterized by vision and colour loss evolving over days and ocular pain intensified by eye movements.[37] However, the widespread pathology can lead to a broad range of symptoms including motor and sensory deficits, pain, bowel and bladder (incontinence) problems, sexual dysfunction, fatigue and cognitive dysfunctions. Although MS is characterized by a range of symptomatologic manifestations, the disease is typically viewed as a motor disease. Up to 90% of people with MS experience motor impairments, which are present across all phases and forms of the disease. Motor impairments are highly disabling and have a considerable impact on the independence and quality of life.[38,39] Given that pwMS are typically diagnosed as young adults and impairments can occur early on, motor disabilities significantly affect personal life choices such as starting a family and employment (Figure 3).[24]

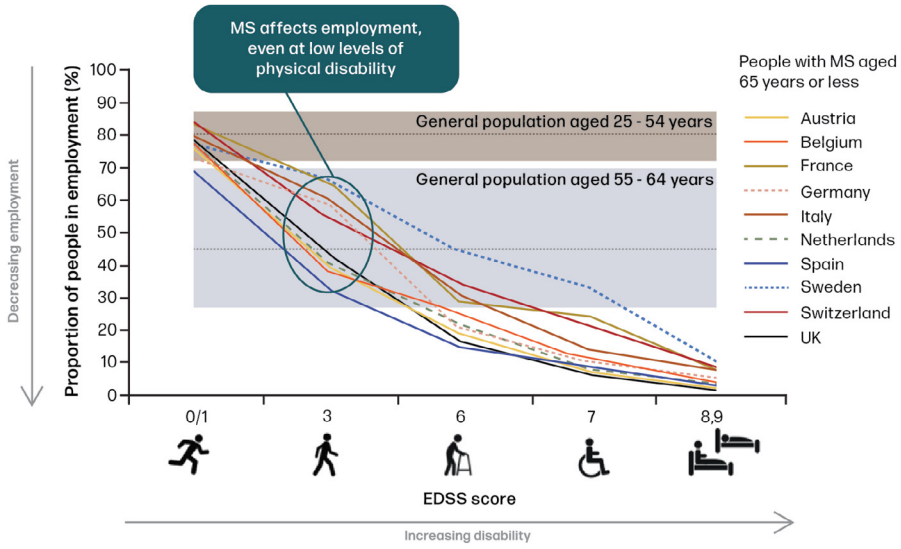


Figure 3. Unemployment in multiple sclerosis. Many people with multiple sclerosis (MS) develop motor impairments due to pathological processes in the brain and spinal cord, which results in unemployment already early on in MS. The proportion of people with MS in employment decreases evidently as disability worsens. Image adapted from Giovannoni et al. 2016.[24]

Functionality of both upper and lower limb limbs can be affected and dysfunctions can present as tremors (postural and action tremor), coordination difficulties, altered sensations (hemisensory), motor weakness, spasticity, balance problems and gait disturbances.[1] Mobility impairments are a typical hallmark of MS and around 80% of pwMS experience difficulties with walking and balance within 10 to 15 years after onset.[40] Upper limb impairments on the other hand are less well recognised, yet disabilities can occur in approximately 80% of pwMS as well.[41] Even though people with MS are highly likely to develop upper and lower limb impairments over the disease course,[42] the degree of dysfunction between limbs correlates only moderately,[43] suggesting that at least partially differential brain processes might underlie.

Treatment

Unfortunately MS is a chronic disease with no known cure, but management is possible. Over the last years, an increasing number of disease modifying treatment options exist for the relapsing-remitting form of MS. These treatments are aimed to modulate or suppress the dysregulated immune system to preserve neurological function, ease symptoms and reduce relapse risk. First-line treatments are prescribed in pwMS that present with mild inflammatory disease activity (high-safety drug), but when proven to be inadequate, second-line treatments are considered (more safety risks). Second-line treatments have proven to be particularly effective in RRMS, mostly by preventing immune cell migration to the brain, but seems to be less successful in the progressive stage of the disease,[13] which is characterized by minimal active demyelinating and neurodegeneration. Treatment

options in the progressive stages of MS are limited. A treatment that not only targets the inflammatory component, but also is neuroprotective and promotes remyelination to slow and ultimately prevent disease activity remains one of the largest challenges.

Current treatment options reduce relapse frequency and slow progression of disabilities, but to develop a therapeutic option that is able to prevent disabilities, we need to better understand the pathological mechanisms that underlie impairments. To improve our understanding, we need to study motor function as well as brain mechanisms underlying these functions. Tools to assess motor functioning and imaging methods to visualize pathology and to study the neuronal mechanisms of motor disability in MS will be discussed in the paragraphs below.

Measuring sensorimotor impairments

There are several ways of assessing motor signs and symptoms in MS ranging from simple tests such as a finger-to-nose test to more comprehensive measures like the Expanded Disability Status Scale (EDSS) and advanced measures of gait using motion capture systems.

The EDSS is a well validated measure of disability in MS. This assessment tool is routinely used in clinic to assess and monitor disabilities and is widely used in MS research as a reflection of overall disability. The EDSS is a rating scale ranging from 0 - 10 with steps of 0.5 that represent higher levels of disability and is based on simple clinical tests and visual evaluation.[44] PwMS with EDSS scores between 0 - 4 show no to mild physical disability, are clinically able to walk and do not require any assistance or walking aid. EDSS scores between 4.0 and 5.5 reveal mild impairment in ambulation and scores equal to or beyond 6 refer to loss of mobility. At the higher end of the scale patients require a walking aid, wheelchair or are restricted to bed (scores beyond 8.0). Even though this rating scale is widely used in clinic and research, it is limited by moderate inter-rater reliability[45] and is heavily weighted to walking ability at the higher ends of the scale.

To overcome these issues, upper and lower limb dysfunction can be examined separately using the 9-Hole Peg Test (9-HPT) and Timed-25 Foot Walk Test (T25FW) respectively. The 9-HPT is a commonly used quantitative arm and hand performance tool that requires a participant to place as quickly as possible nine pegs into nine holes, one at the time, and then to remove these pegs again. This test is done for dominant and non-dominant hand and time in seconds to complete these tests is a reflection of dexterity. The T25FW is a quantitative test to assess leg and mobility function and involves a timed 25 feet walk with outcome measure walking speed. The T25FW is considered to be reliable across a range of disability levels and a difference in scores is considered reflective of a change in real-life activities.[46]

To quantify mobility in pwMS in more detail advanced kinematic analysis technologies that allow for 3-dimensional imaging of gait can be used. Kinematics (study of motion) and kinetics (study of causes of motion) of gait can be assessed using motion capture systems, force plates and electromyography. A previous study in MS has demonstrated the added

value of using these techniques to detect subclinical disease changes not evident based on clinical assessment alone.[47] In early stage MS, pwMS displayed altered timing of ankle muscle activations and ankle angles in absence of clinical disability, no acute relapse or change in EDSS.[47]

Advanced MRI to investigate sensorimotor impairments in multiple sclerosis

Whereas conventional MRI methods are essential for diagnosis and monitoring disease progression in clinic, the correlation between lesions load and clinical impairments is only moderate.[21,22] Stronger relations have been observed with more advanced measures such as brain atrophy,[31-33] but these findings still do not explain the clinical heterogeneity fully. Instead of a focal approach, as the brain is a complex system, to understand behaviour we need to understand how brain regions interact with each other. Relatively recent imaging techniques such as diffusion weighted imaging and functional MRI have made it possibly to study structural and function connectivity between regions and the brain it its entity. The use of these techniques in understanding the complex mechanisms driving motor disability in MS will be discussed in the paragraphs below.

Structural damage within the sensorimotor system in multiple sclerosis

Over the last decade, advanced MRI imaging techniques and analysis methods have been developed and/or optimized, allowing to study the microstructural damage in the brain in great detail. Whereas conventional MRI can image pathology at a macrostructural level, to visualize the microstructural organisation of the WM, newer MRI techniques such as diffusion weighted imaging (DWI) are needed. DWI measures the motion of water molecules, with the preferable direction depended on microstructural restrictions such as cell membranes and axons (Box A). Diffusion MRI is therefore an informative tool to quantify axonal and myelin pathology in MS.

In MS, one of the first models and still a commonly used method to describe and characterize preferential displacement of water molecules to assess microstructural damage is diffusion tensor imaging (DTI). Previous research has shown that DTI measures relate more strongly to disability than conventional measures such as lesions load.[48] Diffusion tensor abnormalities are present from the onset of the disease and are not only observed in lesions, but also measured in the normal appearing WM in both cerebrum and cerebellum.[49] Within the sensorimotor system specifically, the WM integrity of important motor tracts in the brain is damaged including the corticospinal tracts (Figure 4), the main descending motor pathway,[50-52] and the corpus callosum,[53,54] a major WM bundle connecting hemispheres, involved in the control of movements.[55] The microstructural damage within these tracts have been related to clinical disability,[52,53] dexterity[51,54,56] and walking[51].

Even though DTI has been used extensively to assess WM integrity, there are several limitations to consider. Firstly, the DTI model assumes that a brain voxel includes a single fibre population in one orientation, however many voxels may contain partial volume fractions of

distinct fibre populations (Figure 5). Multiple fibre orientations can be detected in up to 90% of WM voxels in the brain.[57] Secondly, metrics derived from a tensor provides a suboptimal estimation of axonal injury as it is insensitive to axonal damage specifically, it contains signal from non-neuronal cells and extra-cellular fluids.[58] Axonal density, dispersion, atrophy and (de)myelination can all contribute to the value of FA, which makes interpretation difficult.[58]

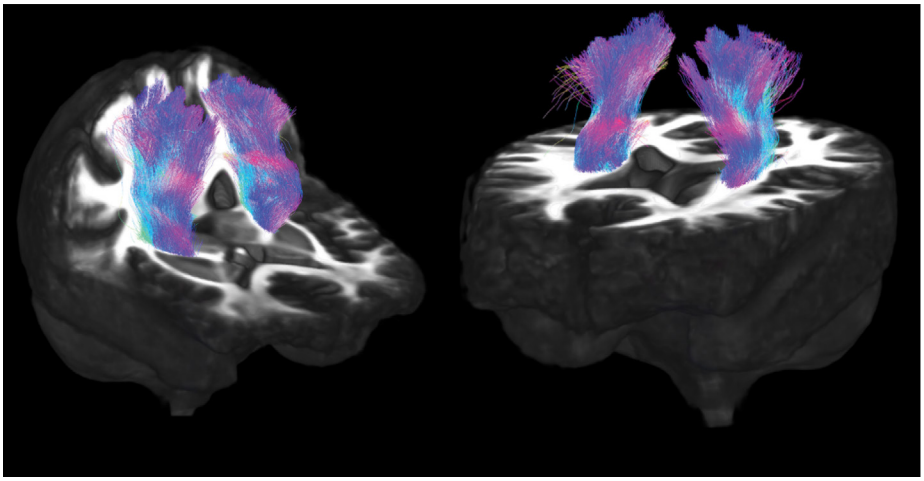


Figure 4. The corticospinal tracts. The corticospinal tract is an important motor pathway that runs from the cerebral cortex to the spinal cord and is involved in the execution of voluntary movements. The motor tracts were derived using diffusion weighted imaging and subsequent analysis method tractography, a technique to delineate white matter tracts in the brain. Whole brain probabilistic tracking was performed first, which involves sampling of diffusion orientations from a large distribution of possible orientations, followed by selection of the corticospinal tract using several inclusion and exclusion regions (described in more detail in Chapter 3.2).

In addition, as neurodegeneration is present from early stages of the disease, there is a high need for early detection with sensitive and specific markers of axonal loss for timely intervention of functional decline and treatments efficacy. To overcome these issues and to investigate sensorimotor axonal loss more specifically more advanced higher order models are needed such as constrained spherical deconvolution. With this method the fibre orientation distribution of multiple fibre bundles within a voxel can be estimated and quantitative measures of the underlying microstructure can be derived (Figure 5).

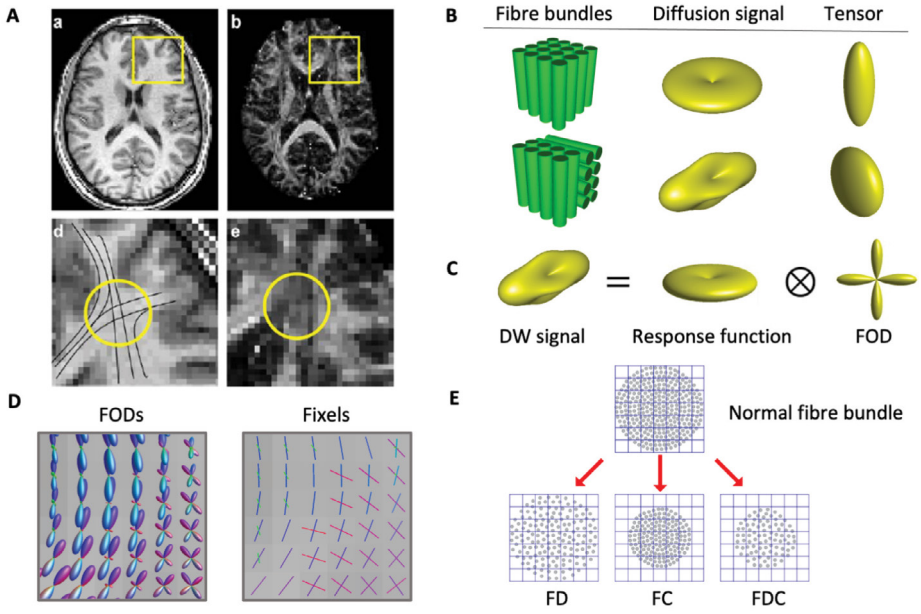


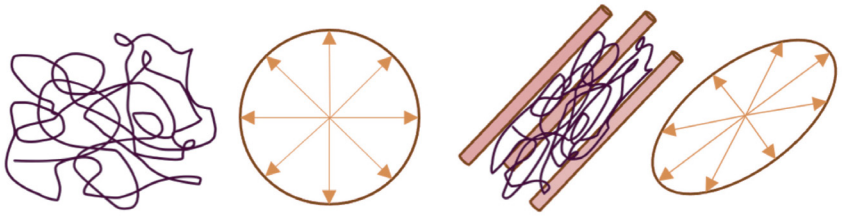
Figure 5. Crossing fibres and diffusion weighted imaging models. A) Partial volume effect and crossing fibres. Image adapted from Assaf et al. 2008.[59] B) Corresponding diffusion signal and diffusion tensor ellipsoid of a signal fibre population (top row) and two crossing fibre populations (bottom row). When fibres cross, the tensor estimated does not reflect the fibre populations and does correspond to the orientation of either population. C) Using constrained spherical deconvolution, the diffusion signal is the convolution between the estimated signal expected for a single-fibre population, i.e. response function, and the fibre orientation distribution (FOD). Image adapted from Raffelt et al 2012.[60] D) The FODs and corresponding fixels, i.e. the specific fibre bundles within a voxel. E) Fixel-specific metrics can be derived, including fibre density (FD), fibre cross-section (FC) or the combination between the two (FDC). The schematic represents the cross sections of fibre bundles. Axons are represented as grey circles and the grid represents imaging voxels.

Functional imaging perspective on sensorimotor disability in multiple sclerosis

Disruptions caused by inflammatory and neurodegenerative processes are highly like to have consequences for brain function and integrity of the system. Brain function can be studied using functional MRI, a non-invasive tool that can assess and quantify neural activation through detection of changes in blood oxygenation (Box B). Functional brain activity can be assessed while performing a specific task (task fMRI). Also, as the brain is a complex system that is highly connected, the communication between regions can be assessed and the brain can be viewed as a network, often studied at rest (resting-state fMRI). Both rest-dependent and task-dependent neural activation has been studied in relation to disabilities in MS, described in the paragraphs below.

Box A – Diffusion Weighted Imaging

Diffusion is driven by the random interaction of water molecules as they collide with one another, known as the Brownian motion. The preferable direction of motion depends on microstructural restrictions such as cell membranes and axons. Axons will effectively restrict movement of water molecules preferentially along the axis of the axon itself. This type of diffusion is known as anisotropic diffusion.



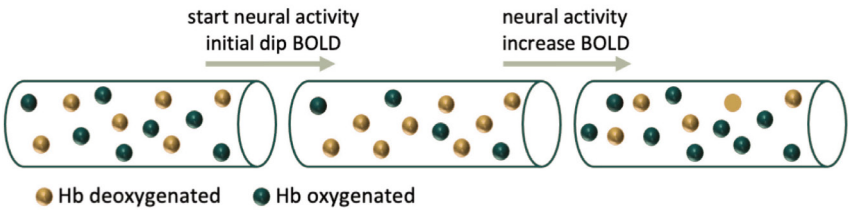
Isotropic diffusion Anisotropic diffusion

The basic diffusion weighted scheme is a spin echo pulse sequence with a gradient magnetic field be pulsed before and after 180 refocusing pulse, known as the diffusion gradients. First, the radiofrequency pulse flip protons in the transverse plane. Secondly, the diffusion gradient before the refocusing pulse results in increased frequency of the spins along the gradient axis. Once the diffusion gradient is turned off the spins will rotate at identical frequency but are phase shifted. Application of the second gradient of equal magnitude and duration but in opposite direction, results in rephasing of spins. Only when spins maintain in exact same position during application of diffusion gradients, spins will rephrase completely. Displacement of spins during dephasing and rephrasing results in incoherent phasing, spins remain out of phase compared to surrounding protons. This displacement of water molecules is the fundament for the signal intensity measured with diffusion MRI. Solid tissues give a strong signal whilst motion of water molecules lead to decrease in signal intensity.

The amount of diffusion (D) is calculated with two images; one with (S) and one without diffusion gradients (S_0), with the following equation; $D = (-\ln(S/S_0))/\text{b-value}$. The factor D describes displacement of protons in all directions and is a property of tissue, but it does not reflect preferential diffusion direction. To detect diffusion accurately, multiple diffusion gradients applied along several directions with different b-values. Higher b-values lead to stronger diffusion weighting but also decrease of signal-to-noise ratio. The magnitude (G), duration (∂), and time interval (Δ) between two diffusion gradients can be described as follows: $\text{b-value} = \gamma^2 G^2 \partial^2 (\Delta - \partial/3)$.

Box B – Functional MRI

The fMRI signal in the brain reflects changes in blood flow in response to neural activity that can be detected using the blood-oxygen-level dependent (BOLD) contrast. Oxygen is transported through the cerebral vascular network and supplied to active neurons by haemoglobin (Hb), an iron-rich protein in red blood cells with primary function to carry and transport oxygen. Upon neural activation, the cerebral blood-flow increases, what results in higher cerebral blood volume and higher oxygen levels, that is delivered to neurons by haemoglobin. Higher oxygen levels, i.e. more bound oxygen to haemoglobin, change the magnetic property of blood that can be detected with fMRI imaging, going from paramagnetic, haemoglobin without oxygen, to diamagnetic properties. The change in BOLD magnetic susceptibilities of haemoglobin and resulting change in T2/T2*-weighted signal is exploited at contrast agent in functional BOLD MRI. Brain regions with more oxy-haemoglobin will have a higher signals and therefore appear brighter on a MRI scan than areas with more deoxy-haemoglobin. Important to note is that the hemodynamic response is located near the location of neural activity and results from blood flow changes directly modulated by neuronal activity, but it is an indirect measure of neural activity.



Functional activation patterns during movements in multiple sclerosis

The functional disturbances within the sensorimotor system underlying motor dysfunctions in MS have been assessed using a task fMRI experiment, which typically involves alternating blocks of the task or stimulus and a control condition, i.e. a period of rest or a task unrelated to the function or behaviour of interest. These alternating blocks of task and control conditions are used to identify activated areas specifically related to the task and to study different activation patterns between groups.

In MS, during a motor task experiment two commonly phenomena are observed, either increased activation in the same regions as healthy controls and/or recruitment of addition regions.[61] In the earlier functional task studies increased brain activity was interpreted as adaptive functional processes in response to structural damage to maintain clinical functions, particularly early on in the disease. One of the earlier motor task fMRI studies showed higher activation in the motor cortices during flexion and extension of the fingers in pwMS and interpreted this change in activation as an adaptive mechanism to limit motor impairment.[62,63] These changes in activation were referred to as functional reorganisation.

The functional counteract against pathological processes was based on the direction of activation, i.e. increased activation was interpreted as adaptive and decreased activation as maladaptive. However, as most studies were based on a cross-sectional design, it is difficult to underpin when changes occur and reoccur and whether cortical reorganisation takes place to delay developments of motor impairments. In addition, the optimal curve and a possible tipping point where functional activation changes are no longer beneficial in sustaining motor disability are not completely understood. Longitudinal studies are limited but warranted to actually comprehend whether cortical plasticity occurs to preserve motor function.

Since these early motor task studies, very little research has emerged on the role of altered brain activation in relation to motor disabilities in MS with the majority of research focussed on either healthy controls[64,65] or used resting-state fMRI to study the motor system.[66,67] While of value in understanding how functional brain networks change in context of sensorimotor progression, resting-state studies are limited by the fact that they do not involve performance of sensorimotor behaviours, and therefore could be difficult to interpret in terms of differences in motor performance. In addition, only a limited number of task fMRI studies have been published investigating lower limb sensorimotor processing,[68,69] possibly due to the complexity of the experimental set-up. Moving your fingers results in less motion than for example tapping your foot and to stabilize the lower limbs to minimize head motion a more complex set-up is needed. In addition, most previous studies used a very simple motor task such as finger tapping or extension and flexion of the fingers,[61-63] which is a poor reflection of daily motor functioning. As such, current literature does not adequately recapitulate the underlying functional activity patterns of upper and lower limb impairments in MS.

Sensorimotor resting-state network in multiple sclerosis

In absence of a task or external input, spontaneous BOLD signal fluctuations can be observed in distinct brain regions and across structurally distributed networks.[70] The significance and motivation to study spontaneous neural activity came from a study showing functional correlation in regions association with motor function, i.e. primary sensorimotor cortices and SMA, in absence of movements.[71] Subsequently, studies have shown correlated temporal patterns of areas involved in motor function, auditory processing and memory, consistent across healthy subjects.[72] Interestingly, these major brain networks at rest closely match with brain activity during activation,[73] suggesting an "active" brain at rest.

Besides mapping brain activity, the connectivity between brain regions is often assessed in absence of a task. The connectivity between two regions reflects how strongly two regions are communicating (Figure 6). In MS, early resting-state studies investigating brain connectivity and plasticity reported altered functional connectivity within several brain networks including the sensorimotor system.[74,75] In early stages of the disease higher functional connectivity was observed and suggestions were made in favour of adaptive

brain plasticity in response to increasing structural damage to sustain clinical functions. Whereas similar suggestions have been made for brain activation in presence of a task or stimuli, different mechanisms are measured, i.e. regional activation strength versus how well brain regions are connected. However, similar to brain activity, over the years it became clear that it was more complex than initially thought. Studies demonstrated not only increased,[75,76] but also reduced connectivity in early disease stages.[77] Looking at the sensorimotor network and physical disability specifically, complex patterns of increased[74,78,79] and decreased[77,80,81] connectivity has been observed, with limited evidence for clinical correlates.[79,80]

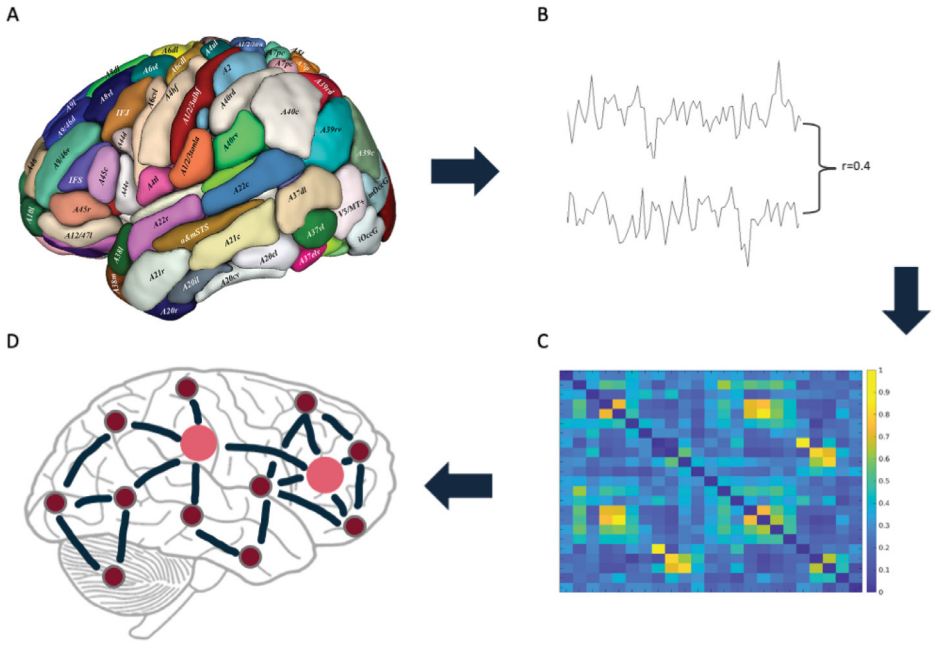


Figure 6. Functional connectivity and network topology. A) The brain can be parcellated in multiple regions of interest. Image adapted from Fan et al. 2016.[82] B) Time series can be extracted from regions of interest and functional connectivity can be calculated using correlation. The correlation strength is a reflection of how strongly two areas are connected. C) Connectivity can be calculated between all regions of interest resulting in a connectivity matrix. The colour indicates the connectivity strength with in blue weak connections and in yellow strongly connected regions. D) A network consists of nodes, representing brain regions, and edges, the functional connections between nodes. From the connectivity matrix in C different network measures can be derived such as efficiency, a measure of information processing in the brain.

The complex patterns of functional connectivity and limited correlates of disability might be explained by variety of reasons including the heterogeneity in terms of group sizes and/or the acquisition and analyses methods. In addition, the conflicting results might be explained by the type of patients investigated, ranging from a specific disease stage, often with high variability in disease duration, or all disease stages. Presumably the type of MS does not explain the complex patterns observed, but motor disability can be better understood by grouping based on level of impairment. Further, most previous functional connectivity studies did not focus on the entire

sensorimotor (Figure 7), rather a specific region or on whole brain network connectivity was used which potentially could overlook relevant areas or include functionally irrelevant areas. Besides differences in study designs, the significant between-subject variability in functional connectivity observed in healthy participants adds to the complexity.[83,84] To be able to compare functional connectivity across subjects the variability should be taken into account.

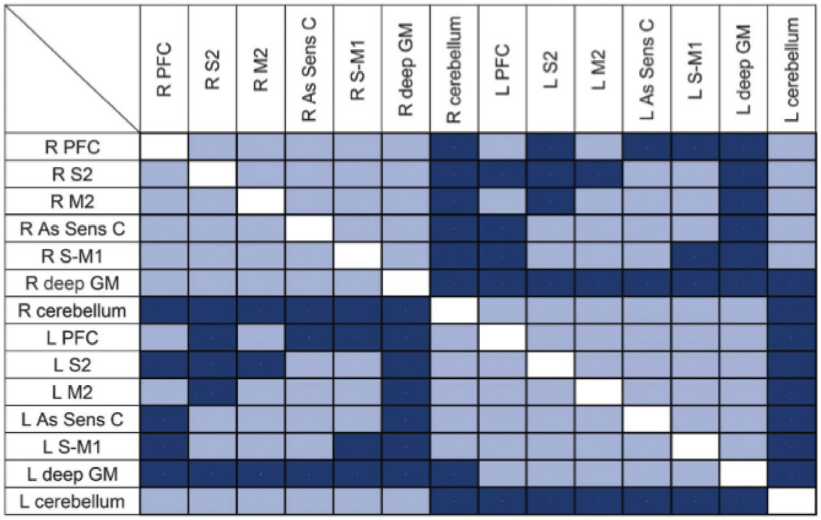


Figure 7. The sensorimotor network. The sensorimotor network contains of multiple cortical and subcortical regions. To delineate white matter tracts and derive the structural connections between these regions a technique called fibre tractography was used. The light blue represents when two regions were structurally connected, in dark blue regions when no tracts between the regions were delineated. The secondary motor area (M2) consists of the supplementary motor regions and premotor area. PFC = prefrontal cortex; S2 = secondary sensory area; As Sens C = associative parietal sensory cortex; S-M1 = primary sensory motor cortex; deep GM = deep gray matter. This image was taken from Pardini et al. 2015.[85]

In addition to investigating the connectivity of spatially separated brain regions, the brain can be studied in its entity. Investigating the system as a whole allows to study the network as a complex system. Simple intuitions from highly complex systems can be derived using a network approach (Figure 6). In MS, previous research has demonstrated altered communication between regions on a network level. Reduced efficiency, a measure of the ability of a region to propagate information to other regions, and decreased local and global efficiency, measures of integration and segregation, were suggested to be clear identifiers of a disrupted network organisation.[86] These changes on a network level emerge from the earliest stage of MS and is consistent with findings from studies using structural imaging modalities.[85,87,88] This evidence for a disrupted network organisation highlights the value of using a network approach in understanding MS pathology. Rather than investigating the connectivity between regions, a network approach could potentially give new insight into the functional mechanisms underlying motor disability in MS.

Ultra-high field imaging

Like network science, over the last years significant progress has been made in ultra-high field imaging (7T). The high signal-to-noise ratio that can be achieved using ultra-high field can be invested in higher spatial resolutions and better contrasts, allowing finer structures to be visualized. In MS, compared to clinical field strengths, previous studies shown the added value of high-field imaging as it improved diagnostic confidence[89] and detection of cortical lesion pathology (Figure 8).[90,91] Besides the ability to image at higher spatial resolutions, a major advantage of high field MRI is functional imaging (Figure 8).

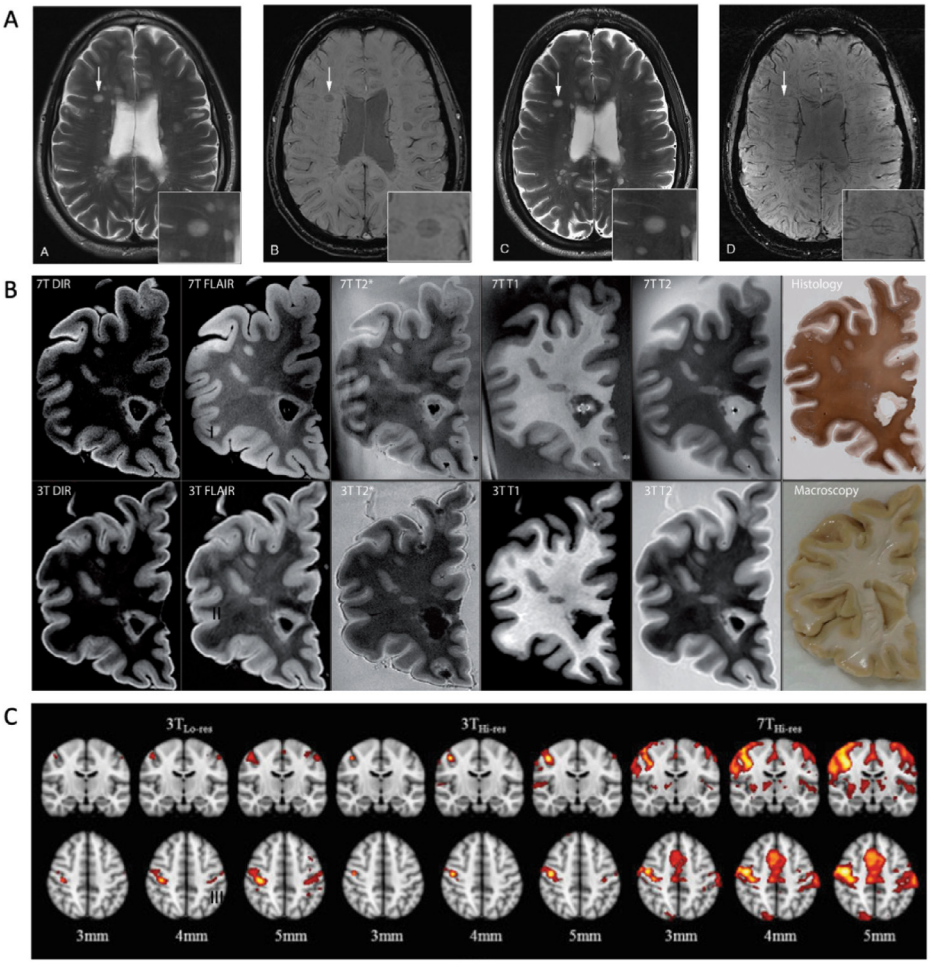


Figure 8. Functional and structural imaging at 3T and 7T in multiple sclerosis. A) Comparison of routine T2-weighted imaging at 3T (A.A) and 7T (A.B) and corresponding axial susceptibility weighted images (SWI) at 3T (A.B) and 7T (A.D).[89] The white arrow indicates the central vessels in MS white matter lesions which are easier and more clearly depicted on 7T compared to 3T SWI. Image adapted from Springer et al. 2016.[89] B) A single brain slice of an MS patient acquired at 7T and 3T using multiple sequences including double inversion recovery (DIR), fluid attenuation inversion recovery (FLAIR), T2*, T1-weighted and T2-weighted.[94] Compared to 3T, higher field imaging resulted in improved quality and grey matter lesion detection. Detected lesion were verified post-mortem. Image adapted from Kilsdonk et al. 2016.[94] C) Compared to 3T, a higher field strength and lower spatial smoothing improves the spatial specificity of connectivity maps and results in the identification of motor cortices. Image adapted from Hale et al. 2010.[92]

The spatial specificity and sensitivity at clinical field strengths is relatively poor. This could possibly impair the detection of subtle changes underlying complex mechanisms underlying impairments such as motor disabilities. Ultra-high field MRI allows to map activations more accurately with minimal spatial smoothing[92] at near anatomical MRI resolution even at reasonable temporal resolution.[93] Functional high-field imaging in MS is still limited used,[92] presumably due to the limited availability, however the rich spatiotemporal content has the potential to detect subtle changes with higher accuracy, potentially improving our understanding of more nuanced motor compensatory mechanisms underlying upper and lower limb motor disability progression in MS.

This thesis: aims and outline

The overarching aim of this thesis was to expand our knowledge of the underlying structural and functional neuronal mechanisms of sensorimotor impairments in pwMS. Despite the evidence of structural damage and functional connectivity disturbances within the sensorimotor system, the clinical heterogeneity is not fully understood. Whereas previous studies often focused on general MS effects, we aimed to study neuronal damage and dysfunction underlying upper and lower limb impairments separately in minimal disabled patients, prior to overt loss of dexterity and mobility, and in patients with more serious disabilities. More sensitive measures of disability as well as more optimal imaging methods were employed to elucidate the association between microstructural damage and brain function and disability in MS.

The overarching research questions of this thesis were

1. Which functional network changes underlie sensorimotor impairments in patients with more severe disabilities, and are these predictive of subsequent progression?
2. In early disability stages, prior to more severe impairments, which functional activation pattern underlie subtle deficits in the upper and lower limbs?
3. In people with MS with minimal physical disabilities, to what extent is axonal loss prevalent in of the major motor pathways of the brain, and does it relate to subtle changes in hand function and walking?

In the first part of this thesis the functional disturbances within the sensorimotor system in a large cohort of pwMS was investigated using a network approach and functional resting-state MRI. First, to get a better understanding of the functional network mechanisms underlying disability severity, in **Chapter 2.1** we aimed to directly compare the functional sensorimotor network efficiency in the presence or absence of overt motor disability. Next, to get a better understanding of disability progression and the mechanisms underlying upper and lower limb specifically, in **Chapter 2.2** baseline network measures were explored as possible predictors of progression of upper and lower limb impairments.

In the second part of this thesis we performed two more focused studies in minimally disabled patients using ultra-high field MRI (7 Tesla) and sensitive measures of motor behaviour derived from force tracking and spatiotemporal measures of gait. With this

dataset we were able to explore the subtle functional changes in brain activity during a complex visuomotor task in relation to subtle changes hand and foot motor performance, addressed in **Chapter 3.1**. In addition, to understand to what extend axonal loss is a feature in pwMS with minimal physical disability, and whether the degree of axon loss relates to subtle changes in upper and lower limb performance, we assessed the white matter integrity within major motor tracts in the brain using novel measures of axonal damage and precise kinematic measures of dexterity and gait, described in **Chapter 3.2**. The key findings of these studies are summarized, integrated and discussed in **Chapter 4**.

References

1. Compston A, Coles A. Multiple sclerosis. Epub ahead of print 2008. DOI: 10.1016/S0140-6736(08)61620-7.

2. O'Gorman C, Lucas R, Taylor B. Environmental risk factors for multiple sclerosis: A review with a focus on molecular mechanisms. International Journal of Molecular Sciences 2012; 13: 11718–11752.

3. Multiple Sclerosis International Federation. Atlas of MS 2013: mapping multiple sclerosis around the world. London, https://www.msif.org/about-us/who-we-are-and-what-we-do/advocacy/atlas/ (2013, accessed 13 July 2020).

4. Lublin FD, Reingold SC, Cohen JA, et al. Defining the clinical course of multiple sclerosis: The 2013 revisions. Neurology 2014; 83: 278–286.

5. Koch M, Kingwell E, Rieckmann P, et al. The natural history of secondary progressive multiple sclerosis. J Neurol Neurosurg Psychiatry 2010; 81: 1039–1043.

6. Lucchinetti CF, Popescu BFG, Bunyan RF, et al. Inflammatory cortical demyelination in early multiple sclerosis. N Engl J Med. Epub ahead of print 2011. DOI: 10.1056/NEJMoa1100648.

7. Haider L, Simeonidou C, Steinberger G, et al. Multiple sclerosis deep grey matter: the relation between demyelination, neurodegeneration, inflammation and iron. J Neurol Neurosurg Psychiatry 2014; 85: 1386–95.

8. Dutta R, Trapp BD. Relapsing and progressive forms of multiple sclerosis: Insights from pathology. Curr Opin Neurol 2014; 27: 271–278.

9. Geurts JJG, Barkhof F. Grey matter pathology in multiple sclerosis. Lancet Neurol 2008; 7: 841–51.

10. Barcellos L. Obesity and Multiple Sclerosis Susceptibility: A Review. J Neurol Neuromedicine 2016; 1: 1–5.

11. Baranzini SE, Oksenberg JR. The Genetics of Multiple Sclerosis: From 0 to 200 in 50 Years. Trends in Genetics 2017; 33: 960–970.

12. Dendrou CA, Fugger L, Friese MA. Immunopathology of MS. Nat Rev Immunol 2015; 15: 545–558.

13. Mahad DH, Trapp BD, Lassmann H. Pathological mechanisms in progressive multiple sclerosis. The Lancet Neurology 2015; 14: 183–193.

14. Stys PK, Zamponi GW, Van Minnen J, et al. Will the real multiple sclerosis please stand up? Nature Reviews Neuroscience 2012; 13: 507–514.

15. Trapp BD, Nave K-A. Multiple Sclerosis: An Immune or Neurodegenerative Disorder? Annu Rev Neurosci. Epub ahead of print 2008. DOI: 10.1146/annurev.neuro.30.051606.094313.

16. Thompson AJ, Banwell BL, Barkhof F, et al. Diagnosis of multiple sclerosis: 2017 revisions of the McDonald criteria. The Lancet Neurology 2018; 17: 162–173.

17. Polman CH, Reingold SC, Banwell B, et al. Diagnostic criteria for multiple sclerosis: 2010 revisions to the McDonald criteria. Ann Neurol 2011; 69: 292–302.

18. Grossman RI, Gonzalez-Scarano F, Atlas SW, et al. Multiple sclerosis. Gadolinium enhancement in MR imaging. Radiology 1986; 161: 721–725.

19. Sahraian MA, Radue EW, Haller S, et al. Black holes in multiple sclerosis: Definition, evolution, and clinical correlations. Acta Neurologica Scandinavica 2010; 122: 1–8.

20. Benedict RHB, Weinstock-Guttman B, Fishman I, et al. Prediction of Neuropsychological Impairment in Multiple Sclerosis: Comparison of Conventional Magnetic Resonance Imaging Measures of Atrophy and Lesion Burden. Arch Neurol 2004; 61: 226–230.

21. Daams M, Steenwijk MD, Wattjes MP, et al. Unraveling the neuroimaging predictors for motor dysfunction in long-standing multiple sclerosis. Neurology 2015; 85: 248–55.

22. Barkhof F. The clinico-radiological paradox in multiple sclerosis revisited. Current Opinion in Neurology 2002; 15: 239–245.

23. De Stefano N, Stromillo ML, Giorgio A, et al. Establishing pathological cut-offs of brain atrophy rates in multiple sclerosis. J Neurol Neurosurg Psychiatry 2016; 87: 93–99.

24. Giovannoni G, Butzkueven H, Dhib-Jalbut S, et al. Brain health: time matters in multiple sclerosis. Multiple Sclerosis and Related Disorders 2016; 9: S5–S48.

25. Fisher E, Lee JC, Nakamura K, et al. Gray matter atrophy in multiple sclerosis: A longitudinal study. Ann Neurol 2008; 64: 255–265.

26. Calabrese M, Magliozzi R, Ciccarelli O, et al. Exploring

the origins of grey matter damage in multiple sclerosis. Nature Reviews Neuroscience 2015; 16: 147–158.

27. Lassmann H. Multiple sclerosis pathology. Cold Spring Harbor Perspectives in Medicine 2018; 8: a028936.

28. Popescu V, Klaver R, Voorn P, et al. What drives MRI-measured cortical atrophy in multiple sclerosis? Mult Scler 2015; 21: 1280–1290.

29. Valsasina P, Aboulwafa M, Preziosa P, et al. Cervical cord T1-weighted hypointense lesions at MR imaging in multiple sclerosis: Relationship to cord atrophy and disability. Radiology 2018; 288: 234–244.

30. Eshaghi A, Marinescu R V, Young AL, et al. Progression of regional grey matter atrophy in multiple sclerosis. Brain 2018; 1665–1677.

31. Calabrese M, Atzori M, Bernardi V, et al. Cortical atrophy is relevant in multiple sclerosis at clinical onset. J Neurol 2007; 254: 1212–1220.

32. De Stefano N, Airas L, Grigoriadis N, et al. Clinical relevance of brain volume measures in multiple sclerosis. CNS Drugs 2014; 28: 147–156.

33. Eshaghi A, Prados F, Brownlee WJ, et al. Deep gray matter volume loss drives disability worsening in multiple sclerosis. Ann Neurol 2018; 83: 210–222.

34. Azevedo CJ, Overton E, Khadka S, et al. Early CNS neurodegeneration in radiologically isolated syndrome. Neurol Neuroimmunol NeuroInflammation 2015; 2: e102.

35. Rashid W, Davies GR, Chard DT, et al. Upper cervical cord area in early relapsing-remitting multiple sclerosis: Cross-sectional study of factors influencing cord size. J Magn Reson Imaging 2006; 23: 473–476.

36. Casserly C, Seyman EE, Alcaide-Leon P, et al. Spinal Cord Atrophy in Multiple Sclerosis: A Systematic Review and Meta-Analysis. J Neuroimaging 2018; 28: 556–586.

37. Toosy AT, Mason DF, Miller DH. Optic neuritis. Lancet Neurol 2014; 13: 83–99.

38. Kierkegaard M, Einarsson U, Gottberg K, et al. The relationship between walking, manual dexterity, cognition and activity/participation in persons with multiple sclerosis. Mult Scler 2012; 18: 639–46.

39. Lamers I, Feys P. Assessing upper limb function in multiple sclerosis. Mult Scler 2014; 20: 775–84.

40. Souza A, Kelleher A, Cooper R, et al. Multiple sclerosis and mobility-related assistive technology: Systematic review of literature. J Rehabil Res Dev 2010; 47: 213–224.

41. Johansson S, Ytterberg C, Claesson IM, et al. High concurrent presence of disability in multiple sclerosis: Associations with perceived health. J Neurol 2007; 254: 767–773.

42. Kister I, Bacon TE, Chamot E, et al. Natural history of multiple sclerosis symptoms. Int J MS Care 2013; 15: 146–158.

43. Coghe G, Corona F, Pilloni G, et al. Is There Any Relationship between Upper and Lower Limb Impairments in People with Multiple Sclerosis? A Kinematic Quantitative Analysis. Epub ahead of print 2019. DOI: 10.1155/2019/9149201.

44. Kurtzke JF. Rating neurologic impairment in multiple sclerosis: an expanded disability status scale (EDSS). Neurology 1983; 33: 1444–52.

45. Hobart J, Freeman J, Thompson A. Kurtzke scales revisited: The application of psychometric methods to clinical intuition. Brain 2000; 123: 1027–1040.

46. Motl RW, Cohen JA, Benedict R, et al. Validity of the timed 25-foot walk as an ambulatory performance outcome measure for multiple sclerosis. Mult Scler J 2017; 23: 704–710.

47. Martin CL, Phillips BA, Kilpatrick TJ, et al. Gait and balance impairment in early multiple sclerosis in the absence of clinical disability. Mult Scler 2006; 12: 620–8.

48. Harrison DM, Shiee N, Bazin PL, et al. Tract-specific quantitative MRI better correlates with disability than conventional MRI in multiple sclerosis. J Neurol 2013; 260: 397–406.

49. Roosendaal SD, Geurts JJG, Vrenken H, et al. Regional DTI differences in multiple sclerosis patients. Neuroimage 2009; 44: 1397–1403.

50. Tallantyre EC, Bø L, Al-Rawashdeh O, et al. Clinico-pathological evidence that axonal loss underlies disability in progressive multiple sclerosis. Mult Scler 2010; 16: 406–411.

51. Hubbard EA, Wetter NC, Sutton BP, et al. Diffusion

tensor imaging of the corticospinal tract and walking performance in multiple sclerosis. J Neurol Sci 2016; 363: 225–231.

52. Tovar-Moll F, Evangelou IE, Chiu AW, et al. Diffuse and Focal Corticospinal Tract Disease and Its Impact on Patient Disability in Multiple Sclerosis. J Neuroimaging 2015; 25: 200–206.

53. Kolasa M, Hakulinen U, Brander A, et al. Diffusion tensor imaging and disability progression in multiple sclerosis: A 4-year follow-up study. Brain Behav; 9. Epub ahead of print 1 January 2019. DOI: 10.1002/brb3.1194.

54. Ozturk A, Smith SA, Gordon-Lipkin EM, et al. MRI of the corpus callosum in multiple sclerosis: Association with disability. Mult Scler 2010; 16: 166–177.

55. Geffen GM, Jones DL, Geffen LB. Interhemispheric control of manual motor activity. Behav Brain Res 1994; 64: 131–140.

56. Kern KC, Sarcona J, Montag M, et al. Corpus callosal diffusivity predicts motor impairment in relapsing-remitting multiple sclerosis: A TBSS and tractography study. Neuroimage 2011; 55: 1169–1177.

57. Jeurissen B, Leemans A, Tournier J-D, et al. Investigating the prevalence of complex fiber configurations in white matter tissue with diffusion magnetic resonance imaging. Hum Brain Mapp 2013; 34: 2747–2766.

58. Beaulieu C. The basis of anisotropic water diffusion in the nervous system - A technical review. NMR in Biomedicine 2002; 15: 435–455.

59. Assaf Y, Pasternak O. Diffusion tensor imaging (DTI)-based white matter mapping in brain research: A review. Journal of Molecular Neuroscience 2008; 34: 51–61.

60. Raffelt D, Tournier JD, Rose S, et al. Apparent Fibre Density: A novel measure for the analysis of diffusion-weighted magnetic resonance images. Neuroimage 2012; 59: 3976–3994.

61. Rocca MA, Colombo B, Falini A, et al. Cortical adaptation in patients with MS: A cross-sectional functional MRI study of disease phenotypes. Lancet Neurol. Epub ahead of print 2005. DOI: 10.1016/S1474-4422(05)70171-X.

62. Reddy H, Narayanan S, Arnoutelis R, et al. Evidence for adaptive functional changes in the cerebral cortex with axonal injury from multiple sclerosis. Brain 2000; 2314–20.

63. Lee M, Reddy H, Johansen-Berg H, et al. The motor cortex shows adaptive functional changes to brain injury from multiple sclerosis. Ann Neurol 2000; 47: 606–613.

64. Alahmadi AAS, Samson RS, Gasston D, et al. Complex motor task associated with non-linear BOLD responses in cerebro-cortical areas and cerebellum. Brain Struct Funct 2016; 221: 2443–2458.

65. Iandolo R, Bellini A, Saiote C, et al. Neural correlates of lower limbs proprioception: An fMRI study of foot position matching. Hum Brain Mapp 2018; 1929–1944.

66. Cordani C, Meani A, Esposito F, et al. Imaging correlates of hand motor performance in multiple sclerosis: A multiparametric structural and functional MRI study. Mult Scler J 2019; 1–12.

67. Bollaert RE, Poe K, Hubbard EA, et al. Associations of functional connectivity and walking performance in multiple sclerosis. Neuropsychologia 2018; 117: 8–12.

68. Ciccarelli O, Toosy AT, Marsden JF, et al. Functional response to active and passive ankle movements with clinical correlations in patients with primary progressive multiple sclerosis. J Neurol 2006; 253: 882–891.

69. Filippi M, Rocca MA, Mezzapesa DM, et al. Simple and complex movement-associated functional MRI changes in patients at presentation with clinically isolated syndromes suggestive of multiple sclerosis. Hum Brain Mapp 2004; 21: 108–117.

70. Fox MD, Raichle ME. Spontaneous fluctuations in brain activity observed with functional magnetic resonance imaging. Nat Rev Neurosci 2007; 8: 700–711.

71. Biswal B, Zerrin Yetkin F, Haughton VM, et al. Functional connectivity in the motor cortex of resting human brain using echo-planar mri. Magn Reson Med 1995; 34: 537–541.

72. Damoiseaux JS, Rombouts SARB, Barkhof F, et al. Consistent resting-state networks across healthy subjects. Proc Natl Acad Sci U S A 2006; 103: 13848–13853.

73. Smith SM, Fox PT, Miller KL, et al. Corre- spondence of the brain's functional architecture during activation and rest. PNAS 2009; 106: 13040–13045.

74. Roosendaal SD, Schoonheim MM, Hulst HE, et al. Resting state networks change in clinically isolated syndrome. Brain 2010; 133: 1612-21.

75. Faivre A, Rico A, Zaaraoui W, et al. Assessing brain connectivity at rest is clinically relevant in early multiple sclerosis. Mult Scler J 2012; 18: 1251-1258.

76. Roosendaal SD, Schoonheim MM, Hulst HE, et al. Resting state networks change in clinically isolated syndrome. Brain 2010; 133: 1612-21.

77. Rocca MA, Valsasina P, Leavitt VM, et al. Functional network connectivity abnormalities in multiple sclerosis: Correlations with disability and cognitive impairment. Mult Scler J 2018; 24: 459–471.

78. Castellazzi G, Debernard L, Melzer TR, et al. Functional connectivity alterations reveal complex mechanisms based on clinical and radiological status in mild relapsing remitting multiple sclerosis. Front Neurol; 9. Epub ahead of print 20 August 2018. DOI: 10.3389/fneur.2018.00690.

79. Dogonowski AM, Siebner HR, Soelberg Sørensen P, et al. Resting-state connectivity of pre-motor cortex reflects disability in multiple sclerosis. Acta Neurol Scand 2013; 128: 328–335.

80. Janssen AL, Boster A, Patterson BA, et al. Resting-state functional connectivity in multiple sclerosis: An examination of group differences and individual differences. Neuropsychologia 2013; 51: 2918–2929.

81. Rocca MA, Valsasina P, Martinelli V, et al. Large-scale neuronal network dysfunction in relapsing-remitting multiple sclerosis. Neurology 2012; 79: 1449-1457.

82. Fan L, Li H, Zhuo J, et al. The Human Brainnetome Atlas: A New Brain Atlas Based on Connectional Architecture. Cereb Cortex. Epub ahead of print 2016. DOI: 10.1093/cercor/bhw157.

83. Chou YH, Panych LP, Dickey CC, et al. Investigation of long-term reproducibility of intrinsic connectivity network mapping: A resting-state fMRI study. Am J Neuroradiol 2012; 33: 833–838.

84. Finn ES, Shen X, Scheinost D, et al. Functional connectome fingerprinting: Identifying individuals using patterns of brain connectivity. Nat Neurosci. Epub ahead of print 2015. DOI: 10.1038/nn.4135.

85. Pardini M, Yaldizli Ö, Sethi V, et al. Motor network efficiency and disability in multiple sclerosis. Neurology 2015; 85: 1115–1122.

86. Liu Y, Wang H, Duan Y, et al. Functional Brain Network Alterations in Clinically Isolated Syndrome and Multiple Sclerosis: A Graph-based Connectome Study. Radiology 2017; 282: 534–541.

87. Fleischer V, Gröger A, Koirala N, et al. Increased structural white and grey matter network connectivity compensates for functional decline in early multiple sclerosis. Mult Scler J 2016; 135245851665150.

88. Shu N, Duan Y, Xia M, et al. Disrupted topological organization of structural and functional brain connectomes in clinically isolated syndrome and multiple sclerosis. Sci Rep; 6. Epub ahead of print 2016. DOI: 10.1038/srep29383.

89. Springer E, Dymerska B, Cardoso PL, et al. Comparison of Routine Brain Imaging at 3 T and 7 T. Invest Radiol 2016; 51: 469–482.

90. Yao B, Hametner S, Van Gelderen P, et al. 7 Tesla magnetic resonance imaging to detect cortical pathology in multiple sclerosis. PLoS One 2014; 9: e108863.

91. Kilsdonk ID, Jonkman LE, Klaver R, et al. Increased cortical grey matter lesion detection in multiple sclerosis with 7 T MRI: A post-mortem verification study. Brain 2016; 139: 1472–1481.

92. Hale JR, Brookes MJ, Hall EL, et al. Comparison of functional connectivity in default mode and sensorimotor networks at 3 and 7T. Magn Reson Mater Physics, Biol Med 2010; 23: 339–349.

93. Yoo PE, John SE, Farquharson S, et al. 7T-fMRI: Faster temporal resolution yields optimal BOLD sensitivity for functional network imaging specifically at high spatial resolution. Neuroimage 2018; 164: 214–229.

94. Kilsdonk ID, Jonkman LE, Klaver R, et al. Increased cortical grey matter lesion detection in multiple sclerosis with 7 T MRI: A post-mortem verification study. Brain 2016; 139: 1472–1481.

CHAPTER 2.1

Increased functional sensorimotor network efficiency relates to disability in multiple sclerosis

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Abstract

Background: Network abnormalities could help explain physical disability in multiple sclerosis (MS), which remains poorly understood.

Objective: This study investigates functional network efficiency changes in the sensorimotor system.

Methods: We included 222 MS patients, divided into low disability (LD, Expanded Disability Status Scale (EDSS) ≤ 3.5, n = 185) and high disability (HD, EDSS ≥ 6, n = 37), and 82 healthy controls (HC). Functional connectivity was assessed between 23 sensorimotor regions. Measures of efficiency were computed and compared between groups using general linear models corrected for age and sex. Binary logistic regression models related disability status to local functional network efficiency (LE), brain volumes and demographics. Functional connectivity patterns of regions important for disability were explored.

Results: HD patients demonstrated significantly higher LE of the left primary somatosensory cortex (S1) and right pallidum compared to LD and HC, and left premotor cortex compared to HC only. The logistic regression model for disability (R^2 = 0.38) included age, deep grey matter volume and left S1 LE. S1 functional connectivity was increased with prefrontal and secondary sensory areas in HD patients, compared to LD and HC.

Conclusion: Clinical disability in MS associates with functional sensorimotor increases in efficiency and connectivity, centred around S1, independent of structural damage.

Introduction

Multiple sclerosis (MS) is a demyelinating and neurodegenerative disorder of the central nervous system leading to disabling sensorimotor impairments.[1] The pathophysiology of neurological dysfunction is complex and not fully understood. Conventional magnetic resonance imaging (MRI) measures of lesion load in brain and spinal cord have shown only modest relations with disability.[2,3] More advanced measures, such as spinal cord[4] and (sub)cortical grey matter (GM) atrophy,[5] relate more strongly to disability, but still do not fully explain clinical heterogeneity, suggesting that additional processes may also be important determinants of disability.

In addition to structural brain changes, functional alterations might be relevant to disability progression. Resting-state functional magnetic resonance imaging (fMRI) has been used to identify changes within sensorimotor network (SMN) from the earliest stages of MS. However, results have been difficult to interpret due to conflicting observations with increased[6–10] and decreased[11–15] functional connectivity (FC), local[14] and widespread connectivity changes,[6,7,12] and limited or conflicting evidence of clinical correlations.[6–9,11–15] In part, differences between studies are likely to reflect differences in cohorts, and limited correlations with outcomes to reflect the inclusion of networks that are not relevant to clinical outcomes that have been assessed.

In this study we assessed the motor system using the concept of network efficiency,[16] a well validated measure of the integration and segregation of information processing in the brain. We assessed functional efficiency in the presence or absence of overt motor disability in a large cohort and investigated the most important correlates of sensorimotor disability in MS.

Methods

Participants

For this study we included 222 patients (age 45.35 ± 10.47, 165 females (74%)) with MS (disease duration 13.60 ± 8.22), part of the Amsterdam MS Cohort[17] and 82 healthy controls (HC, age 45.93 ± 10.79, 52 females (63%)). Inclusion criteria included the presence of relapse-onset MS, availability of disability measurements, either high or low disability severity (see below) and whole brain and cerebellar functional coverage. All patients included in this study were diagnosed with clinically definite MS according to the 2010 revised McDonald criteria,[18] did not experience a relapse in the 2 months prior to the scanning session. This study was approved by the local institutional ethics review board and all participants provided written consent before participation.

Motor disability assessments and group definition

Disability was assessed in patients using the Expanded Disability Status Scale (EDSS).[19] We divided patients into two groups based on walking impairments; that is, low disability (LD, EDSS ≤ 3.5 (no or minimal walking impairment), n = 185, age 43.83 ± 10.12, 135 females) and high disability (HD, EDSS ≥ 6 (unable to walk without aid or assistance), n = 37, age 52.94 ±

8.87, 30 females). Subsequently, the low disability group was reduced into only patients with very minimal disability, EDSS ≤ 2, after which general linear model (GLM) and regression analyses (see below) were repeated.

Imaging data acquisition

All participants were scanned using a 3T-MRI (GE Signa HDxt, Milwaukee, WI) with an 8-channel phased-array head coil. Functional whole-brain resting-state MRI data were acquired with an echo planar imaging sequence (repetition time (TR) = 2200 ms, echo time (TE) = 35 ms, flip angle (FA) = 80°, 3 mm contiguous axial slices, in-plane resolution 3.3 × 3.3 mm^2). For brain volumetric calculations, a three-dimensional (3D) T1-weighted fast spoiled gradient-echo sequence was used (TR = 7.8 ms, TE = 3.0 ms, FA = 12°, inversion time (TI) = 450 ms, 1.0 mm sagittal slices, 0.9 x 0.9 mm^2 in-plane resolution), and a 3D fluid-attenuated inversion-recovery (FLAIR) was acquired to identify white matter (WM) lesions (TR = 8000 ms, TE = 125 ms, TI = 2350 ms, 1.2 mm sagittal slices, 0.98 x 0.98 mm^2 in-plane resolution).

White matter lesion segmentation and brain volume calculations

WM lesions were automatically segmented on FLAIR images using k-nearest neighbor classification with tissue type priors.[20] These images were registered to T1 weighted images and lesions were filled using Lesion Automated Pre-processing.[21] Lesion-filled images were subsequently used to calculate total brain volumes and subcortical volumes using SIENAX and FIRST respectively (https://fsl.fmrib.ox.ac.uk/fsl/). Deep GM (DGM) volumes were subtracted from the total GM volume to calculate cortical GM volume specifically. To account for differences in head size, all brain volumes were normalized using V-scaling factor derived from SIENAX.

Resting-state fMRI pre-processing

Resting-state fMRI pre-processing involved removal of the first two volumes, brain extraction, head motion correction, spatial smoothing with a 5-mm full width at half-maximum Gaussian kernel and high-pass temporal filtering (100 seconds cut off) using the MELODIC pipeline (FSL5). Registration parameters were calculated between fMRI and 3DT1 sequences, using boundary-based registration (BBR), and between 3DT1 and the standard brain using non-linear registration, both of which were inverted to co-register regions of interest (ROIs) to the fMRI sequence (see below). Images were checked for head motion, artefacts and registration errors. Level of motion was calculated based on the average frame-to-frame head motion, as reported previously.[22] The average frame-to-frame head motion did not exceed more than one voxel (3 mm) and did not differ between MS patients and HC (p = 0.36). In addition, voxels without reliable signal caused by echo-planar imaging distortions artefacts or non-brain tissue were excluded using a robust-range based threshold.

Regions of interest

The cortex was segmented using the Brainnetome atlas (http://atlas.brainnetome.org), the cerebellum using the Harvard-Oxford atlas and DGM structures using FIRST. All ROIs were combined to form one atlas, subsequently registered to individual functional scans using inverted BBR parameters and nearest-neighbour interpolation and. assessed on sufficient reliable signal, that is, at least 30% of voxels in at least 90% of all subjects after removing unreliable voxels. This resulted in the removal of orbitofrontal, inferior temporal cortices and nucleus accumbens and a final atlas containing 193 ROIs. Signal intensities within each individual atlas region were averaged for each time-point to form 193 time series and FC was calculated with Pearson correlations, generating a 193 by 193 weighted undirected connectivity matrix. People have very distinct FC profiles,[23] for which we corrected by dividing each connection with average whole-brain FC, enabling us to find disease-related changes in network patterns with relative FC values (Supplementary Figure 1). All negative correlations were set to zero.[22]

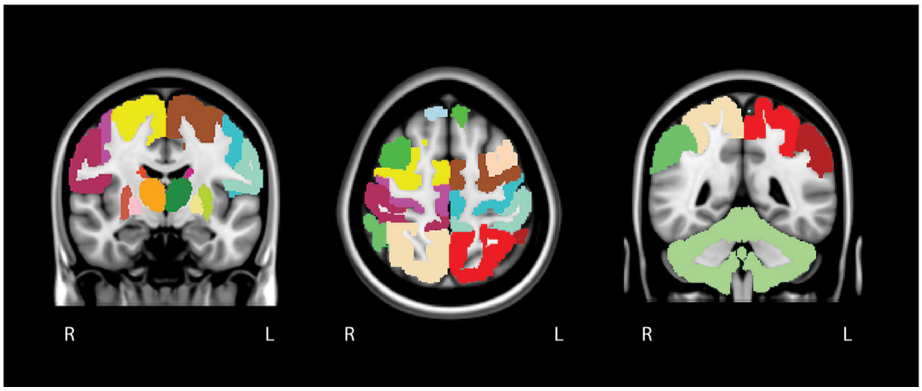


Figure 1. The sensorimotor network. The sensorimotor network was defined based on previous literature and we included 23 cortical and subcortical grey matter areas. The cortical components of the motor network are the primary motor cortex, premotor cortex, supplementary motor area, prefrontal cortex, primary somatosensory cortex, secondary sensory cortex and posterior associative sensory cortex. Each cortical area was subdivided into a right and left region of interest. As for sub-cortical regions, we included the cerebellum and left and right thalamus, caudate nucleus, putamen, and pallidum.

The sensorimotor system

We chose to include a broad range of regions beyond conventional 'typical' motor areas such as primary motor cortex (M1) and somatosensory cortex (S1), based on a previously published approach.[24] This consisted of left and right frontal, parietal and cortical motor regions, deep grey matter areas and the cerebellum, forming a 23 x 23 FC matrix (Figure 1). The cerebellum was included as whole because of limited scan coverage, difficulties in parcellating only motor regions accurately on fMRI and to limit the total number of ROIs. Global efficiency (GE) of the entire SMN and local efficiency (LE) for each ROI was calculated using the Brain Connectivity Toolbox (https://sites.google.com/site/bctnet/). The global efficiency is based on the inverse of the average shortest path length of all individual network links, representing how efficient information flows throughout the entire SMN.

The LE quantifies efficiency on a smaller scale and is related to the clustering coefficient, a network topology characteristic that reflects local processing of information.[16]

Statistical data analyses

All statistical analyses were performed in SPSS 22 (IBM, USA). All measures were checked for normality using the Kolmogorov-Smirnov test and visual inspection of histograms. Lesion volumes were non-normally distributed and log-transformed. Variables with a non-normal distribution were compared with nonparametric tests (Mann-Whitney U test). For normally distributed variables general linear models were used correcting sex and age with Bonferroni-correction over groups; significance after correcting for the number of variables is also reported. To assess the most important correlates of disability, a binary logistic regression model with backward elimination was used including significant network measures, brain and lesion volumes and demographics. GLM and regression analyses repeated including LD patients (EDSS ≤ 2) and HD patients. Areas where LE significantly contributed to the regression model were further explored by examining FC to other sensorimotor areas between groups using GLMs and Spearman correlations were performed between LE and functional system scores (FSS).

Results

Demographics, clinical data, and brain volumes

Demographics, clinical variables and brain volumes are shown in Table 1. The highly disabled group had a significant longer disease duration ($p < 0.001$) and consisted of relatively more secondary progressive MS patients ($p < 0.001$) than the group with low disability. HD patients were significantly older compared to both LD patients ($p < 0.001$) and HC ($p < 0.01$). No differences in handedness were found between patients with high and low disability.

Compared to HC, both MS groups displayed significantly lower whole brain, WM, cortical and DGM volumes ($p < 0.05$). HD patients exhibited more pronounced cortical GM atrophy ($p = 0.001$), DGM atrophy ($p < 0.001$), and whole brain ($p < 0.001$) and WM volume loss ($p < 0.05$) compared to LD patients. In addition, HD patients demonstrated a higher lesion volume compared to LD patients ($p < 0.001$).

Global and local efficiency of the SMN

Global efficiency was not statistically significant. Local efficiency (see Table 2) was higher in the HD group compared to HC in left premotor cortex (p = 0.011), S1 (p = 0.001) and right pallidum (p = 0.044). HD patients showed higher LE in left S1 (p = 0.013) and right pallidum (p = 0.040) compared to LD patients. No differences in efficiency were seen between LD patients and controls. The local efficiency of S1 remains significant after also correcting for the number of variables. These comparisons were not significant when using uncorrected FC matrices.

In addition, when compared to the low disability group based on EDSS ≤ 2, highly disabled patients showed higher GE (p = 0.026) and LE of the left premotor cortex (p = 0.035), M1 (p = 0.011), S1 (p = 0.001), pallidum (p = 0.016) and right M1 (p = 0.033), S1 (p = 0.007), putamen (p = 0.040) and pallidum (p = 0.005). Also here, S1 remains significant after additional correction.

	Healthy controls	Low disability	High disability
Demographics			
• Sex, F/M	52/30	135/50	30/7
• Age, years	45.93 (10.79)	43.83 (10.12)	52.94 (8.87)[a,b]
• Disease duration		12.07 (7.27)	21.23 (8.55)[b]
• Phenotypes (RRMS/SPMS), *n*		178/7	11/26[b]
• EDSS Total[c]		2.5 (0-3.5)	6.5 (6-8)[b]
• FSS Cerebellar[c]		1 (0-3)	3 (0-4)[b]
• FSS Pyramidal[c]		1 (0-3)	3 (2-5)[b]
• FSS Sensory[c]		2 (0-3)	2.5 (1-5)[b]
• FSS Brainstem[c]		0 (0-3)	1 (0-3)[b]
• FSS Visual[c]		0 (0-3)	1 (0-5)[b]
• FSS Bowel and Bladder[c]		0 (0-3)	2 (0-4)[b]
• FSS Cerebral (mental)[c]		1 (0-3)	2 (0-3)[b]
Brain volumes			
• NBV, L	1.51 (0.07)	1.48 (0.07)[a]	1.40 (0.07)[a,b]
• NWMV, L	0.69 (0.03)	0.67 (0.03)[a]	0.65 (0.03)[a,b]
• NCGMV, L	0.78 (0.05)	0.77 (0.05)[a]	0.72 (0.06)[a,b]
• NDGMV, mL	62.85 (3.75)	58.31 (5.80)[a]	51.71 (7.10)[a,b]
• Lesion volume (log), mL		3.88 (0.38)	4.25 (0.40)[b]

Table 1. Demographics, clinical, and MRI characteristics. MRI: magnetic resonance imaging; F: female; M: male; RRMS: relapsing-remitting multiple sclerosis; SPMS: secondary progressive multiple sclerosis; EDSS: Expanded Disability Status Scale; FSS: Functional Systems Scores; NBV: normalized brain volume; NWMV: normalized white matter volume; NCGMV: normalized cortical grey matter volume; NDGMV: normalized deep grey matter volume. All values represent means and standard deviations unless denotes otherwise. [a]Significant difference compared to healthy controls. [b]Significant difference compared to patients with low disability. [c]Median and range.

Sensorimotor regions	HC	LD	HD	*p*-values
Left prefrontal cortex	0.95 (0.11)	0.96 (0.12)	1.00 (0.13)	0.059
Left supplementary motor area	1.09 (0.16)	1.11 (0.20)	1.16 (0.28)	0.391
Left premotor cortex	**1.04 (0.15)**	**1.07 (0.18)**	**1.16 (0.28)[a]**	**0.015**
Left primary motor cortex	1.05 (0.14)	1.08 (0.16)	1.14 (0.20)	0.099
Left primary somatosensory cortex	**1.07 (0.12)**	**1.09 (0.16)**	**1.20 (0.23)[a,b]**	**0.002**
Left secondary sensory cortex	1.06 (0.12)	1.08 (0.14)	1.15 (0.24)	0.051
Left posterior associative sensory cortex	1.07 (0.14)	1.09 (0.16)	1.16 (0.23)	0.172
Left thalamus	1.08 (0.23)	1.11 (0.24)	1.20 (0.37)	0.352
Left caudate nucleus	1.05 (0.21)	1.03 (0.25)	1.12 (0.29)	0.272
Left putamen	1.07 (0.19)	1.05 (0.22)	1.14 (0.28)	0.297
Left pallidum	0.96 (0.20)	0.95 (0.20)	1.08 (0.41)	0.105
Right prefrontal cortex	0.95 (0.16)	0.97 (0.12)	1.01 (0.15)	0.084
Right supplementary motor area	1.10 (0.17)	1.12 (0.19)	1.16 (0.24)	0.556
Right premotor cortex	1.06 (0.15)	1.07 (0.17)	1.14 (0.23)	0.269
Right primary motor cortex	1.05 (0.14)	1.08 (0.16)	1.13 (0.19)	0.188
Right primary somatosensory cortex	1.08 (0.13)	1.10 (0.17)	1.18 (0.21)	0.064
Right secondary sensory cortex	1.07 (0.15)	1.07 (0.15)	1.14 (0.20)	0.395
Right posterior associative sensory cortex	1.07 (0.13)	1.08 (0.17)	1.15 (0.24)	0.257
Right thalamus	1.08 (0.22)	1.10 (0.25)	1.20 (0.34)	0.384
Right caudate nucleus	1.06 (0.20)	1.06 (0.23)	1.11 (0.36)	0.747
Right putamen	1.07 (0.20)	1.06 (0.24)	1.14 (0.27)	0.453
Right pallidum	**0.95 (0.18)**	**0.96 (0.22)**	**1.10 (0.46)[a,b]**	**0.033**
Cerebellum	1.02 (0.20)	1.05 (0.21)	0.96 (0.23)	0.095

Table 2. The mean local efficiency values of each sensorimotor region for each group. HC: healthy controls; LD: patients with low disability; HD: patients with high disability. General linear model was used to compare local efficiency differences between groups and main effects were significant for the left premotor cortex, left primary somatosensory cortex and right pallidum (in bold). [a]After post hoc Bonferroni correction, significant differences were found compared to healthy controls. [b]After post hoc Bonferroni correction, significant differences were found compared to patients with low disability.

Predictors of disability in MS

A final backward binary logistic regression model was created to identify the most important correlates of higher disability (Nagelkerke R^2 = 0.36, Chi-square = 53.56, $p < 0.001$), which included a higher age (Wald = 9.71, $p = 0.002$), lower DGM volume (Wald = 22.19, $p < 0.001$) and higher LE of left S1 (Wald = 6.26, $p = 0.012$). Repeating this regression model using only right-handed patients did not change results. In addition, when repeating this regression model after redefining 'low disability' to EDSS ≤ 2, the same predictors were identified.

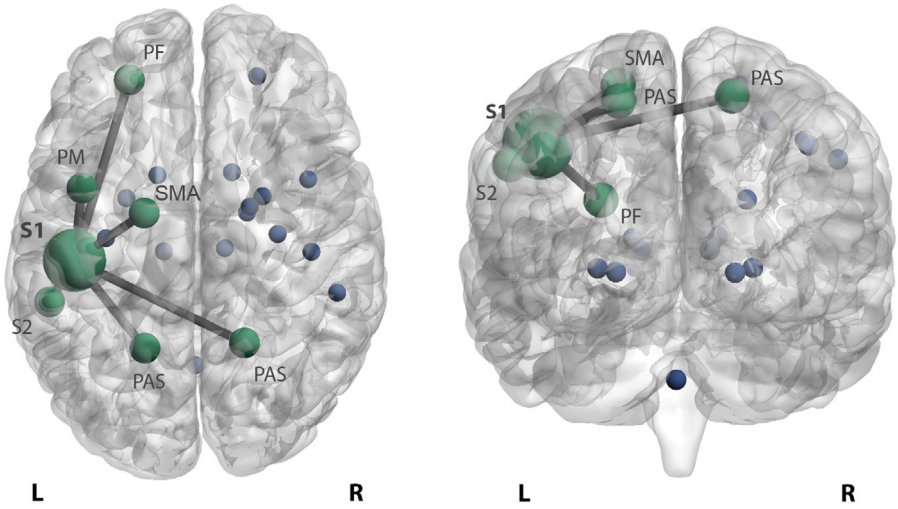


Figure 2. Increased functional connectivity of the somatosensory cortex in highly disabled MS patients. Patients with high disability display higher primary left somatosensory connectivity with the left prefrontal cortex (PF), premotor cortex (PM), secondary sensory cortex (S2) and right and left posterior associative sensory cortex (PAS) compared to patients with low disability and HC. In addition, stronger connectivity with the left supplementary motor area (SMA) was seen in highly disabled patients compared to HC. The stronger connectivity between these areas is reflected by edges between the bigger nodes in green. The blue dots reflect the remaining sensorimotor network regions. Abbreviations: L = left; R = right.

Changes in primary somatosensory FC

HD patients displayed significantly higher FC between left S1 and left prefrontal cortex (p = 0.001 versus HC and p = 0.004 versus LD), premotor cortex (p < 0.001 and p = 0.023), secondary sensory cortex (p = 0.002 and p = 0.011) and right (p = 0.002 and p = 0.026) and left (p < 0.001 and p = 0.007) posterior associative sensory cortex compared to HC and LD patients (Figure 2). In addition, compared only to HC, HD patients displayed higher left S1 connectivity with the left supplementary motor area (SMA) (p = 0.031). In patients with LD, no connectivity changes of the left S1 cortex were found compared to HC. After additional correction, prefrontal, premotor and posterior associative sensory cortices remained significant.

Relations between left S1 changes and clinical functional subsystems

In MS, higher S1 LE significantly correlated with worse pyramidal (r = 0.239, p < 0.001), brainstem (r = 0.210, p = 0.002), sensory (r = 0.211, p = 0.002) FSS and EDSS (r = 0.256, p < 0.001) (Figure 3).

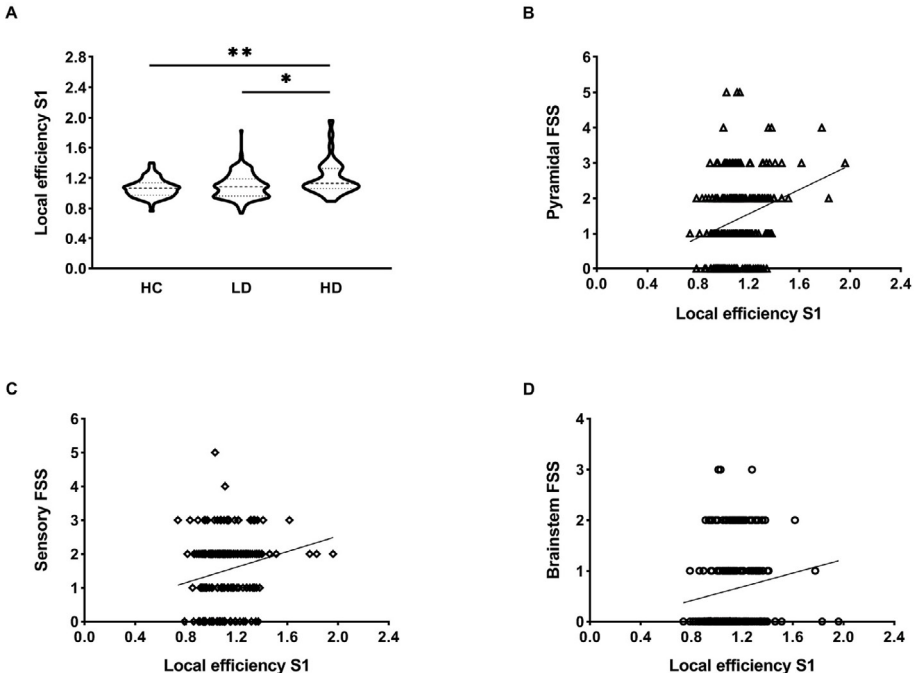


Figure 3. Efficiency of the left primary somatosensory cortex and relations with functional systems scores. (a) Highly disabled MS patients demonstrated significantly higher local efficiency (LE) of S1 compared to both patients with low disability (*, $p < 0.05$) and control subjects (**, $p < 0.01$). The violin plots show the data distributions, that is, the kernel density reflects an estimation of underlying distributed data, and the dashed lines represent the median and the quartiles. (b-d) The scatter plots display significant correlations between LE of left S1 and clinical functional system scores including pyramidal ($r = 0.239$, $p < 0.001$) (b), sensory ($r = 0.211$, $p = 0.002$) (c), and brainstem ($r = 0.210$, $p = 0.002$) (d). Higher LE of S1 significantly correlated with worse pyramidal, brainstem and sensory FSS. HC = healthy controls; LD = patients with low disability; HD = highly disabled patients; S1 = primary somatosensory cortex; FSS = Functional System Scores.

Discussion

In this study, we investigated whether FC and efficiency within the SMN could explain disability in MS and whether specific regions might be particularly affected. Patients with high disability showed extensive SMN changes, mostly centred around S1 as well as the premotor area and pallidum. Patients with minimal disability showed no changes. Increased network efficiency and connectivity of S1 was related to worse disability, even after correcting for structural damage.

S1 and disability in MS

We explicitly identified network changes of S1 that correlated with disability and worse sensory, pyramidal and brainstem FSS (see Figure 3). Impairments in S1 function has shown to be a significant contributor to ineffective motor output and function in other neurological conditions like stroke,[25] but in MS this has not been demonstrated previously. Previous MS literature investigating functional changes has been heterogeneous, identifying global changes that were typically not directly related to disability.[6–8,12,14] One of the first fMRI studies looking at the sensorimotor system investigated connectivity of M1 and showed

decreased interhemispheric connectivity but without any clinical correlates.[14] Instead of one specific area, more recent studies investigated the sensorimotor system using independent component analyses (ICA) and reported either only subcortical changes[8] or global alterations,[6,7,12] but again no significant clinical relations were found.

Previous literature did not clearly link SMN changes to clinical scores, which could possibly be explained by the common exclusion of cerebellar, subcortical, and prefrontal GM structures. These areas were often not included by independent component analysis as 'motor network regions', but considered as a separate network[10,11,13] or part of other networks investigated,[6,12] despite their critical role in sensorimotor functioning and processing. This omission of previous sensorimotor connectivity findings within the framework of motor disability may therefore be the result of an overly simplistic view of network functioning, which may be counteracted by applying whole-network measures such as efficiency. Nonetheless, a connectivity approach was valuable post-hoc, as seen in our data, to show that secondary processing areas, that is, prefrontal cortex, premotor cortex, SMA and secondary sensory cortex, had higher connectivity with S1 (Figure 2), the primary predictor of disability in MS, while primary motor areas (such as M1) did not. These supplementary motor areas play an essential role in the fine balance between somatosensory processing and motor production and altered cortical connectivity might therefore reflect disturbed sensorimotor processing.

Complex patterns of increases versus decreases

Overall, both efficiency and connectivity measures in our study showed increases in disabled patients compared to those with lower disability and HC. Higher sensorimotor connectivity has been found previously in early MS patients with either no or minimal disability,[6,7] frequently interpreted as beneficial functional reorganisation to limit clinical impairment although this remains to be proven.[26] In later stages of MS, increased sensorimotor connectivity has also been observed previously but without clinical relations.[8,10] In our study, no FC changes were observed in patients with low disability, while patients with more severe disability only showed increased FC compared to controls. Decreased connectivity was not seen in our data, but has been observed previously, related to worse disability.[11,13] It was previously suggested that a decrease in connectivity could follow from an initial increase in FC, which remains difficult to prove given the strong lack of longitudinal data.

Our findings indicate that increases in connectivity that are sufficient to alter global motor network efficiency might actually not be related to favourable disability outcomes at all. This is supported by previous studies demonstrating association between increased connectivity and worse cognitive functioning,[27] and disability.[28] In our study, S1 also showed higher FC with several sensorimotor regions, which might be driven by a disruption of mechanisms designed to guide meaningful ascending and/or descending sensorimotor information due to pathological processes in the corticospinal tract. This disruption could result in a loss of inhibition of input reaching S1, resulting in higher connectivity, despite the fact that the information contained within these signals could essentially be noise.

However, such causal claims remain speculative, as the opposite may also be true, that is, that the efficiency and connectivity change could reflect a beneficial mechanism present in patients with severe damage only. As our analyses are limited to cross-sectional, non-directional connectivity measures, future studies should pinpoint how such network changes come to be. The use of empirical computational models might give insight into the change and interaction of functional and structural processes over time.[29]

Structure and function

Our study showed that functional measures provide added value beyond structural damage. Previous studies have shown that higher levels of disability correlate moderately to WM damage, measured by lesion load.[2] Recently, structural network efficiency was shown to explain 58% of disability variation, much more than simply averaging damage.[24] In addition, using an empirical informed model, WM damage in the form of loss of diffusion-based tracts, was shown to drive increased connectivity and network efficiency changes.[29] As such, it would be of high interest to combine structural and functional network measures in the future.

GM damage has also been related to disability, especially DGM and thalamic atrophy.[30] The thalamus has gained considerable interest in MS research as atrophy may precede clinical symptoms,[31] and thalamic atrophy and function strongly correlate to both cognitive decline[27] and disability progression.[5,32] While DGM volume was also a significant correlate of disability in our model, we did not observe significant functional efficiency changes in the thalamus, which is in line with a previous study showing no correlation between thalamic atrophy and thalamic connectivity.[27] We did find an increased efficiency of the pallidum and S1, both areas strongly connected to the thalamus and important in the regulation of movement and sensory processing. The pallidum as a correlate of disability was supported by another recent study.[33] Together, these studies highlight the added value of regional and network-based information.

Future directions

We included areas beyond 'typical' motor regions such as M1 and S1, based on a previous study,[24] but not cognitive regions that could still influence disability or cerebellar subregions. Furthermore, we used relative FC scores due to inter-participant variabilities, that warrant some caution.[23] How best to correct for this variability requires future studies to determine. Even though a recent study showed that cerebral network changes can explain disability beyond spinal cord atrophy,[24] the latter was not included in our study. Furthermore, other graph analytical concepts such as network centrality could provide additional information. Including neurological tests such as the nine-hole peg test could detect other aspects of sensorimotor dysfunction, enabling a more comprehensive way of defining disability. Finally, our approach was based on commonly used 'static' connectivity, that is, efficiency across the entire scan, but unique information may reside in dynamic fluctuations and stability of connectivity patterns.[34,35] Finally, future multimodal longitudinal studies are needed to investigate the order of events leading to the accumulation of disability.

Conclusion

Using an advanced network imaging approach, this study shows that functional changes in the sensorimotor network centred around S1 are associated with high disability, independently from structural damage. Patients with severe disability (aid or assistance required to walk) showed increased local efficiency and connectivity of S1, suggesting that increases in brain network efficiency may be a marker of poorer clinical function.

References

1. Kister I, Bacon TE, Chamot E, et al. Natural history of multiple sclerosis symptoms. Int J MS Care 2013; 15: 146–158.

2. Daams M, Steenwijk MD, Wattjes MP, et al. Unraveling the neuroimaging predictors for motor dysfunction in long-standing multiple sclerosis. Neurology 2015; 85: 248–55.

3. Valsasina P, Aboulwafa M, Preziosa P, et al. Cervical cord T1-weighted hypointense lesions at MR imaging in multiple sclerosis: Relationship to cord atrophy and disability. Radiology 2018; 288: 234–244.

4. Song X, Li D, Qiu Z, et al. Correlation between EDSS scores and cervical spinal cord atrophy at 3T MRI in multiple sclerosis: A systematic review and meta-analysis. Multiple Sclerosis and Related Disorders; 37. Epub ahead of print 1 January 2020. DOI: 10.1016/j.msard.2019.101426.

5. Eshaghi A, Marinescu R V, Young AL, et al. Progression of regional grey matter atrophy in multiple sclerosis. Brain 2018; 1665–1677.

6. Faivre A, Rico A, Zaaraoui W, et al. Assessing brain connectivity at rest is clinically relevant in early multiple sclerosis. Mult Scler J 2012; 18: 1251–1258.

7. Roosendaal SD, Schoonheim MM, Hulst HE, et al. Resting state networks change in clinically isolated syndrome. Brain 2010; 133: 1612–21.

8. Dogonowski A-M, Siebner HR, Sørensen PS, et al. Expanded functional coupling of subcortical nuclei with the motor resting-state network in multiple sclerosis. Mult Scler 2013; 19: 559–66.

9. Dogonowski AM, Siebner HR, Soelberg Sørensen P, et al. Resting-state connectivity of pre-motor cortex reflects disability in multiple sclerosis. Acta Neurol Scand 2013; 128: 328–335.

10. Castellazzi G, Debernard L, Melzer TR, et al. Functional connectivity alterations reveal complex mechanisms based on clinical and radiological status in mild relapsing remitting multiple sclerosis. Front Neurol; 9. Epub ahead of print 20 August 2018. DOI: 10.3389/fneur.2018.00690.

11. Janssen AL, Boster A, Patterson BA, et al. Resting-state functional connectivity in multiple sclerosis: An examination of group differences and individual differences. Neuropsychologia 2013; 51: 2918–2929.

12. Rocca MA, Valsasina P, Martinelli V, et al. Large-scale neuronal network dysfunction in relapsing-remitting multiple sclerosis. Neurology 2012; 79: 1449–1457.

13. Rocca MA, Valsasina P, Leavitt VM, et al. Functional network connectivity abnormalities in multiple sclerosis: Correlations with disability and cognitive impairment. Mult Scler J 2018; 24: 459–471.

14. Lowe MJ, Phillips MD, Lurito JT, et al. Multiple sclerosis: Low-frequency temporal blood oxygen level-dependent fluctuations indicate reduced functional connectivity - Initial results. Radiology 2002; 224: 184–192.

15. Dogonowski AM, Andersen KW, Madsen KH, et al. Multiple sclerosis impairs regional functional connectivity in the cerebellum. NeuroImage Clin 2014; 4: 130–138.

16. Rubinov M, Sporns O. Complex network measures of brain connectivity: Uses and interpretations. Neuroimage. Epub ahead of print 2010. DOI: 10.1016/j.neuroimage.2009.10.003.

17. Eijlers AJC, Wink AM, Meijer KA, et al. Reduced network dynamics on functional MRI signals cognitive impairment in multiple sclerosis. Radiology 2019; 292: 449–457.

18. Polman CH, Reingold SC, Banwell B, et al. Diagnostic criteria for multiple sclerosis: 2010 Revisions to the McDonald criteria. Ann Neurol 2011; 69: 292–302.

19. Kurtzke JF. Rating neurologic impairment in multiple sclerosis: an expanded disability status scale (EDSS). Neurology 1983; 33: 1444–52.

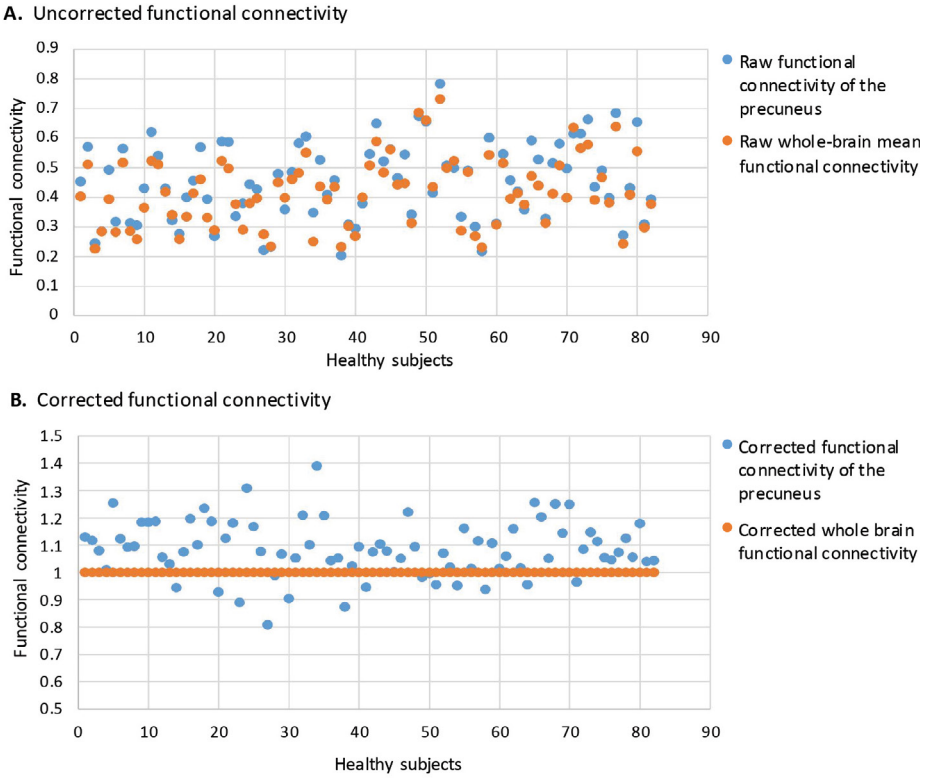
20. Steenwijk MD, Pouwels PJW, Daams M, et al. Accurate white matter lesion segmentation by k nearest neighbor classification with tissue type priors (kNN-TTPs). NeuroImage Clin 2013; 3: 462–469.

21. Chard DT, Jackson JS, Miller DH, et al. Reducing the impact of white matter lesions on automated measures of brain gray and white matter volumes. J Magn Reson Imaging 2010; 32: 223–228.

22. Meijer KA, Eijlers AJC, Geurts JJG, et al. Staging of cortical and deep grey matter functional connectivity

changes in multiple sclerosis. J Neurol Neurosurg Psychiatry 2017; jnnp-2017-316329.
23. Finn ES, Shen X, Scheinost D, et al. Functional connectome fingerprinting: Identifying individuals using patterns of brain connectivity. Nat Neurosci. Epub ahead of print 2015. DOI: 10.1038/nn.4135.
24. Pardini M, Yaldizli Ö, Sethi V, et al. Motor network efficiency and disability in multiple sclerosis. Neurology 2015; 85: 1115–1122.
25. Borich MR, Brodie SM, Gray WA, et al. Understanding the role of the primary somatosensory cortex: Opportunities for rehabilitation. Neuropsychologia 2015; 79: 246–255.
26. Schoonheim MM, Geurts JJG, Barkhof F. The limits of functional reorganization in multiple sclerosis. Neurology, 2010, pp. 1246–1247.
27. Schoonheim MM, Hulst HE, Brandt RB, et al. Thalamus structure and function determine severity of cognitive impairment in multiple sclerosis. Neurology 2015; 84: 776–783.
28. Faivre A, Robinet E, Guye M, et al. Depletion of brain functional connectivity enhancement leads to disability progression in multiple sclerosis: A longitudinal resting-state fMRI study. Mult Scler J 2016; 22: 1695–1708.
29. Tewarie P, Steenwijk MD, Brookes MJ, et al. Explaining the heterogeneity of functional connectivity findings in multiple sclerosis: An empirically informed modeling study. Hum Brain Mapp 2018; 1–8.
30. Eshaghi A, Prados F, Brownlee WJ, et al. Deep gray matter volume loss drives disability worsening in multiple sclerosis. Ann Neurol 2018; 83: 210–222.
31. Azevedo CJ, Overton E, Khadka S, et al. Early CNS neurodegeneration in radiologically isolated syndrome. Neurol Neuroimmunol NeuroInflammation 2015; 2: e102.
32. Rocca MA, Mesaros S, Pagani E, et al. Thalamic damage and long-term progression of disability in multiple sclerosis. Radiology 2010; 257: 463–469.
33. Motl RW, Hubbard EA, Sreekumar N, et al. Pallidal and caudate volumes correlate with walking function in multiple sclerosis. J Neurol Sci 2015; 354: 33–36.
34. Chang C, Glover GH. Time-frequency dynamics of resting-state brain connectivity measured with fMRI. Neuroimage 2010; 50: 81–98.
35. Yaesoubi M, Allen EA, Miller RL, et al. Dynamic coherence analysis of resting fMRI data to jointly capture state-based phase, frequency, and time-domain information. Neuroimage 2015; 120: 133–142.

Supplementary material



Supplementary Figure 1. Inter-subject functional connectivity variability. A) To visualize the variability in functional connectivity (FC) profiles between subjects, for each healthy subject (x-axis) the average whole-brain FC (orange) and the uncorrected FC values (blue) of the precuneus, a major hub in the brain, are plotted on the y-axis. This plot suggests that the precuneus is not a major hub as it is often not higher than the average connectivity of the population. However, compared to each individual whole brain FC, the FC of the precunues is usually higher. B) By dividing each connection by each individual average whole-brain FC, the variability of the hubness of the precuneus is less and can be more readily compared between subjects and groups.

CHAPTER 4

Summary and general discussion

The aim of this thesis was to investigate the mechanism underlying different severities of motor disability in multiple sclerosis (MS). We approached this aim using advanced functional and structural MRI analysis methods in two separate cohorts of people with MS (pwMS).

In the first part of this thesis, we studied the functional sensorimotor network disturbances in a large cohort of pwMS with mild to severe disabilities (Chapter 2). We investigated the functional disturbances within the sensorimotor network in relation to severe disabilities cross-sectionally, and longitudinally in pwMS that progressed or remained stable in terms of upper or lower limbs disabilities. The second cohort included patients with no to minimal clinical impairments and we investigated more subtle motor disabilities (Chapter 3). Here, we used ultra-high field MRI and sensitive measures of motor behaviour to investigate the neuroplasticity of the brain while performing a task and the white matter integrity in relation to subtle changes in motor performance. Together, these four projects aimed to investigate the structural and functional mechanisms as comprehensive as possible across the span of disability in MS. The key questions of this thesis were the following:

1. Which functional network changes underlie sensorimotor impairments in patients with more severe disabilities, and are these predictive of subsequent progression?
2. In early disability stages, prior to more severe impairments, which functional activation pattern underlie subtle deficits in the upper and lower limbs?
3. In people with MS with minimal physical disabilities, to what extent is axonal loss prevalent in of the major motor pathways of the brain, and does it relate to subtle changes in hand function and walking?

In this general discussion, the key findings are summarised and discussed below, as well as my perspective on the directions of future research in MS.

1. Which functional network changes underlie sensorimotor impairments in patients with more severe disabilities, and are these predictive of subsequent progression?

A network perspective to better understand disability in MS?

The sensorimotor network does not only comprise of 'typical' motor regions and tracts such as the primary motor cortex (M1) and corticospinal tracts, but it is a highly intricate system comprised of multiple regions and several pathways that exhibits a high degree of interconnectivity. As MS is characterized by widespread pathological processes, it is highly likely that inflammatory and neurodegenerative processes affect the sensorimotor system. Whereas damage can be focal and sensorimotor functions reside in specialized regions to some degree, the disease is highly heterogeneous, and regions do not act in isolations. The integration and segregation of information flow within and between brain networks lead to complex motor behaviours. As such, the heterogeneity of the disease and the complexity of the system stresses the need to understand the communication within the sensorimotor network. To investigate the functional disturbances within brain networks, resting-state functional MRI has been extensively used. Functional connectivity variations specific to the motor system are present from earliest stages of the disease, but have not clearly been related to clinical symptoms.[1,2] In fact, previous studies have mostly shown complex patterns of increases and decreases in connectivity that have been difficult to interpret. For instance, studies have reported higher connectivity in early stages of MS[1,2], but also reduced connectivity,[3-7] both with conflicting and limited relations to clinical impairments. In addition, methodological issues were frequent, often driven by the incomplete representation of the sensorimotor system. For example, either just the 'typical' motor regions were investigated, such as the primary sensorimotor cortices, overlooking relevant areas, or a whole brain approach was used, potentially including irrelevant regions.

As such, we wanted to take a different approach and study the sensorimotor system in its entirety. We included regions beyond the 'typical' motor regions such as frontal, parietal and subcortical regions, based on previous literature.[8] In addition to investigating the raw connectivity between regions, we studied the system as a whole and investigated how sensorimotor regions worked together as a network. We used the graph theoretic concept of efficiency to study functional disturbances within the sensorimotor network. Network "efficiency" is well validated metric that can be calculated for the entire sensorimotor system (i.e. global efficiency) or for each region separately (i.e. local efficiency). Whereas the global efficiency is based on the average number of steps along the shortest path and reflects efficiency of information transfer at a global level, local efficiency is based on how well-connected neighbour regions, i.e. the ability to integrate information at a local level.

Sensorimotor network abnormalities explain physical disability in MS

Using aforementioned network approach, we were able to demonstrate different patterns of connectivity and efficiency within the sensorimotor system in pwMS with overt disability, while the network of pwMS with no to mild disability remained normal. In pwMS with more serious disabilities (walking difficulties and in need of aid or assistance) three out sensorimotor

regions displayed increased local efficiency including the thalamus, premotor cortex and primary somatosensory (S1) cortex. This binary measure of disability was further deepened by also looking at patients with no to very minimal disability (EDSS < 2) compared to high disability, showing similar results. It should be noted, however, that we did not assess the specific type of disability nor effects of laterality, which might be of interest. The functional changes within the sensorimotor system were especially around the S1 as this region correlated the strongest of disability severity, independent of structural damage. Increased S1 efficiency reflects higher clustering, which means stronger connectivity with neighbour regions. Potentially, increased efficiency of S1 could be caused by pathological processes in the ascending tracts that disrupt sensorimotor information from the periphery to S1, which results in reduced or disturbed signals arriving in S1. Alternatively, damage in descending pathways or in second order pathways connecting parietal or prefrontal cortices to S1 cortices could lead to increased connectivity and efficiency. Changes were observed locally, but not globally. This could be explained by the fact that global and local efficiency metrics are based on different principles. Global efficiency is based on the shortest path length of all individual network links, whereas local efficiency is based on the clustering coefficient which reflects how neighbours of individual nodes are interconnected.

Further, loss of inhibitory inputs might play a role, or the primary sensory cortices might be just more susceptible to damage than other regions in the brain. Also, signals within the system can become more similar due to widespread pathological processes that could lead to higher connectivity and higher efficiency. Unfortunately, no causal claims can be made as a cross-sectional design was used. Nonetheless, given that no network changes were seen in patients without more severe disability, the results from this study suggest that functional network disturbances are clinically disadvantageous in MS.

Sensorimotor network dynamics predict upper and lower limb disability progression

Next, we were interested in studying functional alterations to the sensorimotor network that were related to upper or lower limb impairments specifically and their value in explaining progression of disability. We used a longitudinal design to assess disability progression over 5 years and 24 pwMS were classified as upper limb progressing and 67 as lower limb progressing. Interestingly, only 21 patients converted to SPMS, which indicates that the current definition of SPMS does not overlap our current definition of progression on lower or upper limb motor function. This could indicate that our clinical definition of conversion to SPMS was more conservative than our research definition of progression, although it should be noted that we did not take relapses into account. Further, in addition to measurements of network efficiency across the entire functional scan (i.e. static approach), we also studied fluctuations over time (i.e. network dynamics). As static measurements are not sensitive to the fundamental dynamic nature of both brain and behaviour,[9,10] studying temporal fluctuations of brain connectivity and networks is imperative, but limited research. Whereas a static approach is commonly used in MS research, studying the temporal fluctuations has only been recently validated, mostly focused on cognition.[11,12] In relation to physical disability,

we observed increased network efficiency dynamics and connectivity at baseline in pwMS who subsequently progress. The dynamics of the thalamus and supplementary motor area were independent predictors of upper and lower limb disability progression respectively. The higher dynamics could reflect a network that is unstable and therefore unable to perform as needed. Alternatively, higher network efficiency dynamics could be an adaptive response to ongoing pathology, what might reach have reach a threshold after which dynamics reduces and subsequent progression of disability. In cognitively impaired patients reduced centrality dynamics was observed in the same group of pwMS.[11] Lower centrality dynamics reflects a less fluctuating network that might be interpreted as a network that is 'stuck', what could lead to cognitive dysfunctions. However, from these results it is not clear how these different network metrics and disability and cognition potentially relate to each other. Nonetheless, our results suggest that high fluctuations in sensorimotor network efficiency is a sign of imminent progression of upper and lower limb impairments.

Summary Chapter 2

To conclude, from the studies in Chapters 2.1 and 2.2 we have learned that functional disturbances within the sensorimotor network are more prominent in pwMS who develop more serious motor disabilities. Our results show that while static efficiency increases relate to more severe disability cross-sectionally, dynamic efficiency increases were able to predict imminent clinical progression. These findings highlight the added value of using a network approach and studying the temporal features of a system in understanding the mechanisms underlying disability in MS.

2. In early disability stages, prior to more severe impairments, which functional activation pattern underlie subtle deficits in the upper and lower limbs?

Whereas in aforementioned studies we assessed functional sensorimotor network disturbances in pwMS with severe motor impairments, next were interested in investigating the functional mechanisms underlying more subtle impairments in early stages of disability. We were able to detect changes in brain activity and subtle changes in motor performance in minimally disabled pwMS using a complex visually guided force-matching task and ultra-high field imaging. Compared to healthy controls, during lower limb force tracking, pwMS displayed a delayed response to the task cue and more erroneous movements, as well as lower activity in regions involved in visuomotor integration including cerebellar, occipital and superior parietal areas. Lower activity within these visuomotor regions correlated with worse disability and more structural damage. During upper limb movements, pwMS displayed lower inferior occipital cortical activation, but interestingly we did not observe any correlation with sensorimotor disability or structural damage and any between-group differences in task performance. Together these results suggest that upper limb function is preserved in pwMS with no to minimal impairments and that partially divergent functional mechanisms might underlie upper and lower disabilities.

The neuronal mechanisms and pathways of upper and lower limb movements in brain and spinal cord are highly complicated and are not fully understood. Compared to walking, upper limb movements are more complex, have a higher degree of freedom and are less repetitive and thus less spinal cord processing dependent. Possibly it is easier to adapt upper limb motor functioning due to higher dependence on brain function. The disassociation between upper and lower limb motor performance in MS is in line with previous studies reporting moderate relation between walking and upper limb function[13] and lower limb impairments early in the disease.[14-16] As for brain activity, studies investigating the lower limbs in limited, either in clinically isolated syndrome[17] or primary progressive MS[18]. Studies investigating hand function often used simple motor task at clinical field strength in a different population of pwMS.[19-22] These approaches could have impaired the detection of subtle changes and more nuanced motor mechanisms underlying impairments in both upper and lower limbs, particularly in early disability. From our study we have learned that motor control impairments in MS are related to dysfunctions in visuomotor integration. Furthermore, ultra-high field imaging during complex upper and lower limb force tracking can reveal subtle impairments in movement and brain activation and differentiate upper and lower limb impairments in minimally disabled MS.

3. In people with MS with minimal physical disabilities, to what extent is axonal loss prevalent in of the major motor pathways of the brain, and does it relate to subtle changes in hand function and walking?

In pwMS with no to minimal clinical impairments substantial axonal loss was observed within major sensorimotor tracts, despite minimal focal inflammatory demyelination lesions within the tract, the classical pathological hallmark of MS. Unlike conventional diffusion tensor imaging measures, we used novel axonal markers associated with specific fibre bundles, even in voxels that contain multiple fibre populations. The combination of recent advances in diffusion MRI and signal modelling enabled us to derive markers of axonal loss including fibre bundle density, indicative of diffuse axonal loss, and fibre cross-section, a marker of tract atrophy. The greatest effects were observed at a macrostructural level in the cross-section of the fibre bundle in all three tracts: corticospinal tract, interhemispheric sensorimotor tract, and cerebello-thalamic tracts. The degree of axonal fibre loss was associated with more erroneous hand movements and altered spatiotemporal gait patterns (shorter stance, smaller step width and prolonged double support).

To examine the added value of using a fixel-based approach in understanding disease pathology and the relation to structural damage elsewhere in the system, we examined the correlations between conventional brain volumetrics and fibre specific damage. The sensorimotor tracts were relatively devoid of focal inflammatory demyelination lesions, with less than 21% overlap with the tracts of interest in only up to 29% of pwMS. A moderate correlation was observed between whole brain lesion load and tract damage. While we cannot infer causality, this might suggest that the field of injury driving axonal loss is widespread rather than focal. Potentially, focal damage in other pathways or regions

connecting to primary sensorimotor cortices could lead to trans-synaptic degeneration of the fibres in the sensorimotor tracts. In addition, Wallerian or retrograde axonal degeneration resulting from spinal cord injury could be a contributing factor to the WM damage within the corticospinal tracts. However, within the cervical cord, an area where most corticospinal tracts terminate[23] and lesions are commonly located (59%),[24] we observed no atrophy and no correlation between cervical lesion load and axonal damage in the motor tracts. This might argue against spinal cord lesions contributing to motor tract damage in the brain. In addition, early in the disease spinal cord lesions are often asymptomatic[25] and do not seem to predict disability progression[26]. Further, we observed grey matter atrophy of S1 and thalamus, structures containing neurons whose axons comprise significant proportions of the motor tracts, which correlated with axonal loss in the corticospinal tracts.

Further, from our results it appears that axonal loss in minimally disabled patients is rather non-specific as substantial axonal damage was observed throughout all three motor pathways. However, within the corticospinal tracts we observed that the lower limb tracts were affected to a slightly greater extent than tracts related to upper limb movements. Also, a larger proportion of tracts originating from S1 compared to M1 were damaged. Therefore, it is reasonable to believe that some parts of the sensorimotor network might be more vulnerable to MS pathology and potentially sensory processes might be affected to a greater extent, leading to changes in hand function and walking patterns.

Summary Chapter 3

To conclude, we have gained insight into the changes in functional activation patterns and microstructural damage in relation to subtle changes in upper and lower limb function in pwMS with no to minimal motor impairments. We found subtle changes in brain activity and subtle impairments in movements during lower limb force tracking and preserved upper limb function. In addition, substantial axonal loss was observed within major sensorimotor tracts in the brain, despite relatively few white matter lesions, associated with subtle impairments in hand function and walking.

Observations brought forward in this thesis: bringing it all together

A key role for the primary sensorimotor cortices in disability progression in MS?

Considering all the results of this thesis, of the primary sensorimotor regions S1 was frequently implicated to play a role in disability in MS. In pwMS with minimal disabilities we observed significant S1 atrophy, but M1 was not atrophic, and S1 tracts were affected to a greater extent than axons descending from M1 (Chapter 3.2). As S1 is highly connected to other cortical and subcortical regions, S1 atrophy and tract damage early in disability potentially leads to functional activation changes in other regions such as cerebellar and superior parietal areas as observed during upper and lower limb movements (Chapter 3.1). While in early stages of disability sensory regions and tracts could be more vulnerable to structural damage, subsequently in more severe disabilities, functional alterations within S1 were observed, i.e. S1 efficiency and dynamics predicted disability severity (Chapters 2.1 and

2.2). In more severe disability stages, axonal loss within the ascending pathways might have exceeded a threshold leading to functional disturbances in S1 as well, that consequently leads to progression of motor impairments. However, from our results we cannot infer any causality in disturbances within the primary sensorimotor regions and ascending and descending tracts. To understand the order of events and to unravel the complex relation between structure and function, multimodal approaches and a longitudinal design are needed. In addition, focussing solely on S1 might be too simplistic as the motor system is an intricate network comprised of multiple pathways and a high degree of interconnectivity, a network approach could be useful.

What is the role of the thalamus in MS?

Besides the primary sensorimotor cortices, our results indicate a significant role for the thalamus in disability. The thalamus is an important hub in the brain and is widely connected to other (sub)cortical regions. In minimally disabled pwMS we observed thalamic atrophy which is in line with previous studies showing that thalamic atrophy precedes clinical symptoms in MS[27] and progresses rapidly throughout the disease with strong correlation to physical disability[28] and cognitive dysfunction.[29] In addition, we observed functional disturbances of the thalamus, i.e. the functional efficiency of the thalamus fluctuated more (i.e. higher dynamics) in more severe disability stages. Increased network dynamics is in line with previous research which shown higher thalamic functional connecitivity[29] and increased centrality[30] related to clinical symptoms in MS. Whereas most previous studies focused on either cognition or physical disabilities, motor and cognitive dysfunctions are related to each other[31] possibly due to the integrative role of the thalamus in the entire system. In this thesis, the assessment tools used, and the sensorimotor network studied are highly likely to not only involve pure motor function but also comprise cognitive aspects. For example, we used the 9-HPT to assess hand dexterity, but cognitive aspects are likely to be involved in this test as 9-PHT performance was found to relate to cognition functioning.[32] In addition, the sensorimotor network studied did not comprise of pure motor regions, but also included regions involved in cognition function such as the frontal cortex, deep grey matter structures and the cerebellum. As the brain is a complex system, using a more holistic approach and studying cognition and physical disability together might lead to better understanding of the mechanisms underlying symptoms in MS. Nonetheless, regional information may also be useful, as regions such as the thalamus are a central hub that is highly responsible for the network to perform as needed.

Upper and lower limb impairments in MS

In pwMS with minimal clinical disability, we observed worse motor performance during lower limb movements but preserved upper limb function (Chapter 3.1). In more severe disease stages both upper and lower limb disabilities progressed over 5 years, but more patients displayed a decline in lower limb than upper limb function (65% vs 11%) (Chapter 2.2). These findings suggest that differential functional mechanisms might underlie upper and lower limb disabilities in MS, possibly with the lower limbs being affected earlier in the

disease and more severely. However, even though we did not observe significant between-group differences in pwMS with mild impairments, pwMS did perform slightly worse during upper limb force tracking including more erroneous and delayed responses that correlated with more severe damage within major sensorimotor tracts (Chapter 3.2). These findings suggest that upper limb dysfunction is already present early on, in line with findings of previous studies,[16,33] but were possibly not detected properly and more sensitive tools are needed. Similarly, in later stages, the assessment tools might not have been sensitive enough used to detect upper limb progression accurately.

Alternatively, upper limb disabilities might emerge slightly later and/or more mildly than lower limb impairments and are not clinically present until more severe stages of the disease. We studied either pwMS with minimal motor impairments or pwMS with more serious disabilities, however we did not focus on the group in between. Taking into account that the clinical course of MS is highly heterogenous, studying pwMS with moderate impairments potentially gives more insights into the emerge of upper and lower limb impairments in MS.

Future directions

From clinical rating scales to more sensitive measures of upper and lower functioning

In minimally disabled pwMS we were able to detect subtle lower limb impairments including more erroneous movement and a delayed response during force tracking and altered patterns of gait. We did not observe any changes in upper limb function using the visually guided force-matching fMRI task. Possibly, hand dysfunction emerges in later stages or we were unable to detect subtle dysfunction with the assessment tool used. Even though the task involved complex visuomotor integration and reflects daily motor performance more closely than simple fMRI tasks such as finger tapping, it is limited due to restricted movements possible in the scanner. However, detecting subtle changes in upper limb function early on disease is not only useful in research to understand evolution of disease progression, but is potentially important in clinic as well in order to optimize treatment strategies as early as possibly to delay or prevent progression of disabilities.

The gait patterns and behavioural performance measures acquired during visuomotor fMRI task were assessed in minimally disabled pwMS only but might be informative in more severe disability stages as well. In pwMS with more serious motor disabilities, we used the EDSS, 9-HPT and the T25FW to assess disability severity and the progression of upper and lower limb impairments. Whereas these tests are relatively easy to perform and quick to implement, evaluation is not very detailed. The EDSS has limited reliability,[34] is predominantly sensitive to lower limb impairments particularly at the higher ends of the scale. In MS, the 9-HPT is recommended as a golden standard for measuring fine dextrous manual movements.[35] However impaired dexterity can be accompanied by reductions in upper limb strength, altered hand sensation,[33,36] tremors,[37,38] changes in vibration,[39] abnormal tactile sensibility,[36] and impaired coordination[39]. Unfortunately currently there is no single outcome measure that covers all upper limb related disabilities in MS.[40] Similarly, the T25FW essentially measures walking speed in a straight line, but does not capture other aspects of gait including balance, turning, endurance and subtle changes in walking that require advanced technologies.[14,41]

To detect subtle changes in walking and assess the spatiotemporal aspect of the gait cycle 3-dimensional video tracking gait analyses can be used. While laboratory gait analysis has many advantages over simple physical assessments, this method requires a complex experimental set up with a dedicated laboratory and advanced motion capture systems, sensor embedded walkways and force platforms. To obtain more simple but accurate measures of walking performance, wearable wireless sensors can be used.[41,42] Wearable sensors can be placed on different parts of the body to measure acceleration, orientation, velocity and gravitational forces or under the foot to detect changes in pressure and force, both used to asses walking and balance.[41] Sensors give less detailed information but are easy to use, lower in costs and can potentially improve monitoring of lower limb impairment in clinic as well as in daily life between visits to detect changes as early as possible for optimal treatment.[41]

Whereas wearable sensors could potentially be very useful, current assessment tools such as the 9-HPT and T25FW might be used more optimal. In Chapter 2.2 we defined progression based on > 20% decline in performance, but we lacked healthy control data and were therefore unable to define who was disabled at baseline. This would be useful not only in research but also in clinic to define whether upper or lower limb function is actually impaired. Currently, treatment strategies are optimized when a relapse occurs, or when progression is clinically measured, but when deviations from normal are detected earlier, potentially treatment strategies can be adapted or formulated to prevent functional decline.

Clinical and ultra-high field imaging

Our ultra-high field imaging findings have highlighted the added value of 7T MRI in understanding the underlying neuronal mechanisms of motor impairments in MS. We studied the sensorimotor system using 3T imaging in a large heterogenous group of pwMS and performed a more focussed study in minimally disabled pwMS using ultra-high field MRI. Whereas 3T MRI scanners are widely available, ultra-high field MRI allows to image the brain in more detail and with higher accuracy. The high signal-to-noise ratio that can be achieved with ultra-high field allows the visualisation of finer structures and has shown to improve detection of cortical grey matter pathology[43,44] and diagnosis[45] in MS. Besides better structural imaging, functional imaging at ultra-high field has major advantages over 3T imaging. The higher signal-to-noise can be invested in higher spatiotemporal resolutions and better contrasts improving the statistical power and modelling of the signal, which allows to detect subtle functional activation changes with higher specificity.[46,47] At lower field strengths the SNR is enhanced commonly by using large spatial smoothing kernels to improve inter-subject alignments, but also leads to incorrect estimation of true localisation and consequently to type-I errors.[48,49] Ultra-high field imaging comes with the challenge of worse magnetic field inhomogeneities. Signal dropout and warping is particularly observed around the inferior temporal and orbitofrontal cortices. B0 and B1 field inhomogeneities leading to signal distortions area particularly seen with diffusion weighted imaging.

In this thesis, 7T imaging allowed us to detect subtle changes in brain activation and microstructural damage in absence of lesion pathology in relation to subtle changes in upper and lower limb function pwMS with no to minimal impairments. The clinical application of ultra-high field imaging is still limited, but these findings highlight the added value of 7T to detect subtle changes early in the disease. This could be useful in monitoring and predicting the emergence of motor impairments and could lead to more effective treatment strategies to delay or prevent significant neurological decline.

Multi-model approaches

Given that the brain is an incredibly complex and highly connected system, it is very likely that the sensorimotor network overlaps and is connected with other brain networks involved in for example cognitive function. A more holistic approach could be useful in understanding the mechanisms underlying disabilities in MS. In addition, brain function and structure are often studied separately, but likely the interplay between different modalities underlies disabilities in MS. Spatially structural and functional resting-state network largely resemble each other.[50] However whereas the structure of the brain can be considered relatively fixed, brain function is more dynamic. Damage to one pathway could result in several variations in functional activation patterns. Potentially the change in coupling between structural and functional networks lead to motor deficits. A multi-model approach and longitudinal studies are needed to decode the complex coupling between structure and function and the evolution of sensorimotor impairments. In addition, current developments in artificial intelligence now allow for a reliable modelling of brain function. Such models have been used to simulate progression in MS, by simulating structural network disconnection due to lesions, and evaluating functional consequences to the network.[51] These explorations have shown that such structural damage initially induces increased functional connectivity, which transitions to reduced functional connectivity in later stages. In addition, a sudden loss of network efficiency (a "network collapse") was identified during this transition towards reduced connectivity. These approaches could be useful to further our understanding of the network changes we have observed in this thesis, as well as their relevance for motor dysfunction as well as cognitive impairment.

From advanced MRI methods to imaging biomarkers in clinic

Whereas we have learned a great deal about motor disability in MS using advanced imaging methods, the translation of these methods to clinic is challenging. Whereas network measures and/or markers of axonal loss might be useful in monitoring and predicting the emergence of mobility and dexterity impairments in MS, the studies were largely exploratory involving complex analysis and were based on a group rather than the individual. While these techniques might be quite far away from clinical monitoring, phase II trials could be are a good way to translate these techniques because secondary endpoints often involve advanced imaging.[52,53] In addition, as MS is highly heterogenous, there is an urgent need to predict the individual trajectories, but current knowledge is limited on how to do this. To capture the clinical heterogeneity and understand the individual brain mechanisms underlying each disability

course, ultra-high field imaging could be useful because of the high sensitivity which allows to image the brain at high spatiotemporal resolutions. In addition, machine-learning could be helpful in building better predictive models of individual disease courses, however the application in neuroscience is still relatively new and these techniques require large and variable datasets to train.

Histopathology of axonal loss and network disturbances

We have learned a great deal about motor disability in MS using advanced imaging methods, however the histopathology underlying the MRI changes were not studied. We have inferred lower fibre cross-section and fibre density in major motor tracts as axonal loss. Even though this was based on solid theory and high-quality imaging, histopathological validation is needed. Histopathological measures of axonal density and cross-section would be valuable for accurately interpreting fibre bundle changes in pwMS. For instance, recent work has investigated the underlying substrate of diffusion changes in cortical lesions,[54] while another study focused on diffusion changes within white matter tract connected to damaged cortical areas.[55] The use of our techniques based on fixel analyses and 7T imaging could provide even more information. Modelling the fibre orientation distributions and deriving fibre specific metrics of axonal density and cross-section, as well as the high spatial resolutions acquired with 7T imaging,[56,57] might allow for more accurate estimation of pathology and diffusivity. In addition, our network effects could also be investigated in more detail using histopathological studies, as was done recently.[58] This study focussed on the relation between neuronal size and axonal density and structural network topology measures in pwMS. Smaller neurons and lower density at a micro-scale related to a higher macro-scale clustering coefficient, a measure of network segregation. Fibre length, a measure of integration, related to neuronal size, i.e. longer fibers were associated with larger neurons.[58] These relations not only show the potential underlying pathological mechanisms of network changes, but also suggests that a network approach might be informative in understanding the neuroaxonal pathological processes in MS. Even though these initial findings are promising, the histopathological basis of particularly functional network changes remains poorly understood. Improving our understanding of the pathology processes underlying these network disturbances might lead to novel treatment targets.

Key Findings

The key findings of this thesis can be summarised as follows:

1. Functional network efficiency disturbances and severe disabilities

- Out of 222 MS patients, 185 patients displayed minimal impairments and 37 patients presented with more serious disabilities
- In pwMS with no to mild disability the sensorimotor network remained normal
- PwMS with severe disabilities displayed increased sensorimotor network efficiency
- Functional changes in highly disabled patients were centred around the somatosensory cortex (S1)
- S1 connectivity was increased with prefrontal and secondary sensory areas
- Out of 214 pwMS, 24 showed upper and 67 showed lower limb progression
- Progression was predicted by increased network efficiency dynamics
- No change in static network measures observed in progressing compared non-progressing patients
- Thalamic and supplementary motor area dynamics predicted upper and lower limb disability progression respectively
- Functional network effects were predictors of progression independent of structural damage

2. Functional task activation patterns and motor behaviour in minimally disabled pwMS

- Differential functional activity patterns and motor performance were observed for hand and foot impairments in patients with no to minimal disabilities
- PwMS displayed delayed and more erroneous lower limb force tracking compared to healthy controls
- Lower cerebellar, occipital and superior parietal cortical activation was observed during lower limb movements in pwMS compared to controls
- Despite no differences in upper limb task performance, pwMS displayed lower activation in the inferior occipital cortex
- Ultra-high field during a complex visuomotor fMRI task is useful to detect subtle impairments in movement and subtle changes in brain activity

3. Axonal loss and subtle changes in hand function and walking

- Minimally disabled pwMS displayed substantial axonal loss within major sensorimotor tracts, despite relatively few focal lesions
- The degree of axonal loss associated with worse hand function and altered patterns of walking

Future studies are needed to further investigate

Network disturbances and progression of sensorimotor impairments

- Can we unravel whether network metrics initially increase till a certain threshold is reached, after which connectivity and network efficiency decreases, leading to progression of motor disability in MS?
- What is the interplay between structural damage and functional changes in ascending (S1) and descending (M1) pathways in relation to motor disability progression?
- Can we more precisely unravel the drivers of disability using a multi-modal approach?
- What are the histopathological substrates of the disturbed network patterns observed in this thesis?
- Will the integration of a network perspective and more sensitive measures (i.e. high field imaging + gait analysis) help to further unravel disability progression?

The use and development of sensitive tests and ultra-high field imaging

- Could axonal fibre metrics provide useful markers for emerging sensorimotor network dysfunction in early disease stages?
- Could decline in motor function be detected earlier in clinic with more sensitive measures, and would this be useful to optimize treatment strategies to prevent neurological decline?
- Could ultra-high field MRI markers be useful in the motoring and predicting decline in hand and foot function?

Progression of upper and lower limb disabilities

- Are more sensitive measures needed to detect upper limb disabilities early on?
- We studied either minimally or severely impaired pwMS, but can we further elucidate the neural mechanism underlying upper and lower limb disability progression by studying the sensorimotor network of mildly impaired pwMS?
- What are the network disturbances of upper and lower limb impairments over a longer time frame, and would this help to unravel the order of impairments?

References

1. Faivre A, Rico A, Zaaraoui W, et al. Assessing brain connectivity at rest is clinically relevant in early multiple sclerosis. Mult Scler J 2012; 18: 1251–1258.

2. Roosendaal SD, Schoonheim MM, Hulst HE, et al. Resting state networks change in clinically isolated syndrome. Brain 2010; 133: 1612–21.

3. Janssen AL, Boster A, Patterson BA, et al. Resting-state functional connectivity in multiple sclerosis: An examination of group differences and individual differences. Neuropsychologia 2013; 51: 2918–2929.

4. Rocca MA, Valsasina P, Martinelli V, et al. Large-scale neuronal network dysfunction in relapsing-remitting multiple sclerosis. Neurology 2012; 79: 1449–1457.

5. Rocca MA, Valsasina P, Leavitt VM, et al. Functional network connectivity abnormalities in multiple sclerosis: Correlations with disability and cognitive impairment. Mult Scler J 2018; 24: 459–471.

6. Lowe MJ, Phillips MD, Lurito JT, et al. Multiple sclerosis: Low-frequency temporal blood oxygen level-dependent fluctuations indicate reduced functional connectivity - Initial results. Radiology 2002; 224: 184–192.

7. Dogonowski AM, Andersen KW, Madsen KH, et al. Multiple sclerosis impairs regional functional connectivity in the cerebellum. NeuroImage Clin 2014; 4: 130–138.

8. Mayhew SD, Porcaro C, Tecchio F, et al. fMRI characterisation of widespread brain networks relevant for behavioural variability in fine hand motor control with and without visual feedback. Neuroimage 2017; 148: 330–342.

9. Bolton TAW, Morgenroth E, Preti MG, et al. Tapping into Multi-Faceted Human Behavior and Psychopathology Using fMRI Brain Dynamics. Trends in Neurosciences. Epub ahead of print 15 July 2020. DOI: 10.1016/j.tins.2020.06.005.

10. Fox MD, Snyder AZ, Vincent JL, et al. The human brain is intrinsically organized into dynamic, anticorrelated functional networks. Proc Natl Acad Sci U S A 2005; 102: 9673–9678.

11. Eijlers AJC, Wink AM, Meijer KA, et al. Reduced network dynamics on functional MRI signals cognitive impairment in multiple sclerosis. Radiology 2019; 292: 449–457.

12. d'Ambrosio A, Valsasina P, Gallo A, et al. Reduced dynamics of functional connectivity and cognitive impairment in multiple sclerosis. Mult Scler J 2019; 1–13.

13. Coghe G, Corona F, Pilloni G, et al. Is There Any Relationship between Upper and Lower Limb Impairments in People with Multiple Sclerosis? A Kinematic Quantitative Analysis. Mult Scler Int 2019; 2019: 1–6.

14. Martin CL, Phillips BA, Kilpatrick TJ, et al. Gait and balance impairment in early multiple sclerosis in the absence of clinical disability. Mult Scler 2006; 12: 620–8.

15. Benedetti MG, Piperno R, Simoncini L, et al. Gait abnormalities in minimally impaired multiple sclerosis patients. Mult Scler 1999; 5: 363–368.

16. Kister I, Bacon TE, Chamot E, et al. Natural history of multiple sclerosis symptoms. Int J MS Care 2013; 15: 146–158.

17. Filippi M, Rocca MA, Mezzapesa DM, et al. Simple and complex movement-associated functional MRI changes in patients at presentation with clinically isolated syndromes suggestive of multiple sclerosis. Hum Brain Mapp 2004; 21: 108–117.

18. Ciccarelli O, Toosy AT, Marsden JF, et al. Functional response to active and passive ankle movements with clinical correlations in patients with primary progressive multiple sclerosis. J Neurol 2006; 253: 882–891.

19. Rocca MA, Colombo B, Falini A, et al. Cortical adaptation in patients with MS: A cross-sectional functional MRI study of disease phenotypes. Lancet Neurol. Epub ahead of print 2005. DOI: 10.1016/S1474-4422(05)70171-X.

20. Rocca MA, Gavazzi C, Mezzapesa DM, et al. A functional magnetic resonance imaging study of patients with secondary progressive multiple sclerosis. Neuroimage 2003; 19: 1770–1777.

21. Pantano P, Mainero C, Domenico Iannetti G, et al.

Cortical motor reorganization after a single clinical attack of multiple sclerosis. Neuroimage 2002; 17: 1837-1843.
22. Mezzapesa DM, Rocca MA, Rodegher M, et al. Functional cortical changes of the sensorimotor network are associated with clinical recovery in multiple sclerosis. Hum Brain Mapp 2008; 29: 562-573.
23. Patestas M, Gartner LP. A textbook of neuroanatomy, 2nd Edition. 2nd ed. Wiley-Blackwell, 2016.
24. Weier K, Mazraeh J, Naegelin Y, et al. Biplanar MRI for the assessment of the spinal cord in multiple sclerosis. Mult Scler J 2012; 18: 1560-1569.
25. Granella F, Tsantes E, Graziuso S, et al. Spinal cord lesions are frequently asymptomatic in relapsing-remitting multiple sclerosis: a retrospective MRI survey. J Neurol 2019; 266: 3031-3037.
26. Dekker I, Sombekke M, Witte B, et al. Asymptomatic spinal cord lesions do not predict the time to disability in patients with early multiple sclerosis. Mult Scler J 2017; 23: 153 -156.
27. Azevedo CJ, Overton E, Khadka S, et al. Early CNS neurodegeneration in radiologically isolated syndrome. Neurol Neuroimmunol NeuroInflammation 2015; 2: e102.
28. Eshaghi A, Prados F, Brownlee WJ, et al. Deep gray matter volume loss drives disability worsening in multiple sclerosis. Ann Neurol 2018; 83: 210-222.
29. Schoonheim MM, Hulst HE, Brandt RB, et al. Thalamus structure and function determine severity of cognitive impairment in multiple sclerosis. Neurology 2015; 84: 776-783.
30. Eijlers AJC, Meijer KA, Wassenaar TM, et al. Increased default-mode network centrality in cognitively impaired multiple sclerosis patients. Neurology 2017; 88: 952-960.
31. Slavkovic S, Golubovic S, Vojnovic M, et al. Influence of cognitive and motor abilities on the level of current functioning in people with multiple sclerosis. Zdr Varst 2019; 58: 54-61.
32. Yozbatiran N, Baskurt F, Baskurt Z, et al. Motor assessment of upper extremity function and its relation with fatigue, cognitive function and quality of life in multiple sclerosis patients. J Neurol Sci 2006; 246: 117-122.
33. Bertoni R, Lamers I, Chen CC, et al. Unilateral and bilateral upper limb dysfunction at body functions, activity and participation levels in people with multiple sclerosis. Mult Scler 2015; 21: 1566-1574.
34. Hobart J, Freeman J, Thompson A. Kurtzke scales revisited: The application of psychometric methods to clinical intuition. Brain 2000; 123: 1027-1040.
35. Feys P, Lamers I, Francis G, et al. The Nine-Hole Peg Test as a manual dexterity performance measure for multiple sclerosis. Multiple Sclerosis 2017; 23: 711-720.
36. Guclu-Gunduz A, Citaker S, Nazliel B, et al. Upper extremity function and its relation with hand sensation and upper extremity strength in patients with multiple sclerosis. NeuroRehabilitation 2012; 30: 369-374.
37. McCreary JK, Rogers JA, Forwell SJ. Upper limb intention tremor in multiple sclerosis: An evidence-based review of assessment and treatment. Int J MS Care 2018; 20: 211-223.
38. Labiano-Fontcuberta A, Benito-León J. Understanding Tremor in Multiple Sclerosis: Prevalence, Pathological Anatomy, and Pharmacological and Surgical Approaches to Treatment. Tremor and Other Hyperkinetic Movements; 2. Epub ahead of print 2012. DOI: 10.7916/D8Z60MR3.
39. Svendsen KB, Jensen TS, Hansen HJ, et al. Sensory function and quality of life in patients with multiple sclerosis and pain. Pain 2005; 114: 473-481.
40. Lamers I, Feys P. Assessing upper limb function in multiple sclerosis. Mult Scler 2014; 20: 775-84.
41. Shanahan CJ, Boonstra FMC, Cofré Lizama LE, et al. Technologies for Advanced Gait and Balance Assessments in People with Multiple Sclerosis. Front Neurol; 8. Epub ahead of print 2 February 2018. DOI: 10.3389/fneur.2017.00708.
42. Janshen L, Santuz A, Ekizos A, et al. Fuzziness of muscle synergies in patients with multiple sclerosis indicates increased robustness of motor control during walking. Sci Rep; 10. Epub ahead of print 2020.

DOI: 10.1038/s41598-020-63788-w.
43. Yao B, Hametner S, Van Gelderen P, et al. 7 Tesla magnetic resonance imaging to detect cortical pathology in multiple sclerosis. PLoS One 2014; 9: e108863.
44. Kilsdonk ID, Jonkman LE, Klaver R, et al. Increased cortical grey matter lesion detection in multiple sclerosis with 7 T MRI: A post-mortem verification study. Brain 2016; 139: 1472-1481.
45. Springer E, Dymerska B, Cardoso PL, et al. Comparison of Routine Brain Imaging at 3 T and 7 T. Invest Radiol 2016; 51: 469–482.
46. Constable RT, Spencer DD. Repetition time in echo planar functional MRI. Magn Reson Med 2001; 46: 748–755.
47. Yoo PE, John SE, Farquharson S, et al. 7T-fMRI: Faster temporal resolution yields optimal BOLD sensitivity for functional network imaging specifically at high spatial resolution. Neuroimage 2018; 164: 214–229.
48. Heidemann RM, Ivanov D, Trampel R, et al. Isotropic submillimeter fMRI in the human brain at 7 T: Combining reduced field-of-view imaging and partially parallel acquisitions. Magn Reson Med 2012; 68: 1506–1516.
49. Sacchet MD, Knutson B. Spatial smoothing systematically biases the localization of reward-related brain activity. Neuroimage 2013; 66: 270–277.
50. Van Den Heuvel MP, Mandl RCW, Kahn RS, et al. Functionally linked resting-state networks reflect the underlying structural connectivity architecture of the human brain. Hum Brain Mapp 2009; 30: 3127–3141.
51. Tewarie P, Steenwijk MD, Brookes MJ, et al. Explaining the heterogeneity of functional connectivity findings in multiple sclerosis: An empirically informed modeling study. Hum Brain Mapp 2018; 39: 2541–2548.
52. Cortese R, Collorone S, Ciccarelli O, et al. Advances in brain imaging in multiple sclerosis. Therapeutic Advances in Neurological Disorders; 12. Epub ahead of print 1 June 2019. DOI: 10.1177/1756286419859722.
53. Tur C, Moccia M, Barkhof F, et al. Assessing treatment outcomes in multiple sclerosis trials and in the clinical setting. Nature Reviews Neurology 2018; 14: 75–93.
54. Preziosa P, Kiljan S, Steenwijk MD, et al. Axonal degeneration as substrate of fractional anisotropy abnormalities in multiple sclerosis cortex. DOI: 10.1093/brain/awz160.
55. Kiljan S, Preziosa P, Jonkman LE, et al. Cortical axonal loss is associated with both gray matter demyelination and white matter tract pathology in progressive multiple sclerosis: Evidence from a combined MRI-histopathology study. Mult Scler J 2020; 135245852091897.
56. Jonkman LE, Klaver R, Fleysher L, et al. The substrate of increased cortical FA in MS: A 7T post-mortem MRI and histopathology study. Mult Scler 2016; 22: 1804–1811.
57. Granberg T, Fan Q, Treaba CA, et al. In vivo characterization of cortical and white matter neuroaxonal pathology in early multiple sclerosis. Brain 2017; 140: 2912–2926.
58. Kiljan S, Meijer KA, Steenwijk MD, et al. Structural network topology relates to tissue properties in multiple sclerosis. J Neurol 2019; 266: 212–222.

CHAPTER 5

Appendices

Abstract

Multiple sclerosis (MS) is an autoimmune disorder of the brain and spinal cord, and the most common cause of neurological disability in young adults. The presentation of MS is highly heterogeneous with an unknown aetiology and no known cure, presenting as inflammation, demyelination and axonal injury/loss. MS pathology is disseminated throughout the central nervous system leading to a broad range of symptoms including cognitive dysfunctions, bowel and bladder problems, fatigue, sensory disturbances and difficulties with walking and balance.

Up to 90% of people with MS experience motor impairments that significantly worsen with increasing disease severity, and which can affect both the upper and lower limbs. Motor impairments are often highly debilitating, ranging from muscle weakness, coordination loss, tremors to spasticity. However, while the pathophysiological mechanisms underpinning motor impairments in MS have been widely studied, they are not currently well understood. This is particularly true for early disease, a time when personalised treatment strategies can be formulated and will have maximal effect, preventing future deterioration and accumulation of disability. Consequently, there is an urgent need to understand the pathophysiology underlying motor impairments in MS and elucidate the microstructural and functional changes that occur at their earliest manifestation.

To this end, we investigated the pathophysiology of motor impairments associated with dexterity and mobility in people with MS using advanced magnetic resonance imaging (MRI). Our investigations included functional resting-state (Chapters 2.1 and 2.2) and task (Chapter 3.1) MRI, diffusion weighted imaging (Chapter 3.2) and ultra-high field MRI (Chapters 3.1 and 3.2). Our findings consistently demonstrated a clear link between the development of motor impairments and alterations in the structure/function of the sensorimotor system, a system responsible for the integration of sensory information with motor processing in order to facilitate and maintain movement. Specifically, studying the sensorimotor system in its entirety using network analyses in a large cohort of people with MS, we identified functional disturbances within the sensorimotor system of patients with serious disabilities (Chapter 2.1), with disturbances particularly predictive of future progression of upper and lower limb impairments (Chapter 2.2). Further, using high resolution ultra-high field MRI and measures of motor behaviour in a cohort of patients with minimal motor impairments (Chapters 3.1 and 3.2), we similarly found a link between changes in the function and microstructure of the sensorimotor system and the presence of subtle impairments in hand function and walking.

These findings provide evidence for the role of the sensorimotor network in the development of motor impairments. Potentially, the sensorimotor network might be central to the development of motor impairments in MS and represent a useful target for the development of imaging biomarkers for use in treatment development as well as understanding and monitoring the evolution of motor impairments. From this and subsequent work, it is hoped that this knowledge will lead to more effective treatments and management of patients, alleviating the burden of these impairments.

Samenvatting

Wat is multiple sclerose?

Multiple sclerose (MS) is een progressieve neurodegeneratieve en neuro-inflammatoire aandoening van de hersenen en het ruggenmerg, ook wel het centrale zenuwstelsel genoemd. MS is een van de meest voorkomende oorzaken van invaliditeit bij jonge mensen en wordt vaak gediagnostiseerd tussen de 20 en 40 jaar. Hoe MS begint is nog onbekend en welke symptomen wanneer optreden verschilt per persoon. Een breed scala aan symptomen kunnen zich voordoen zoals cognitieve stoornissen, spraakproblemen, vermoeidheid, sensorische stoornissen en problemen met lopen en evenwicht. Hoewel MS wordt gekenmerkt door al deze verschillende symptomen, wordt de ziekte doorgaans gezien als een motorische aandoening omdat deze symptomen het meest zichtbaar zijn. Rond 90% van de mensen met MS ervaart motorische beperkingen van zowel de armen als de benen die aanzienlijk kunnen verslechteren naarmate de ziekte ernstiger wordt. Vrouwen worden vaker gediagnosticeerd met MS dan mannen en er zijn aanwijzingen dat bepaalde genetische en omgevingsfactoren kunnen bedragen. Ondanks deze aanwijzingen begrijpen we nog niet waarom de ene persoon MS krijgt en de ander niet.

De meest voorkomende vorm van MS is relapsing-remitting MS (RRMS). Dit type MS wordt gekenmerkt door terugvallen die minstens 24 uur duren. Deze terugvallen worden ook wel relapses, exacerbaties of schubs genoemd. Tijdens een terugval worden de symptomen ernstiger, maar binnen een aantal weken tot maanden beleven de meeste mensen met RRMS een herstelperiode (remissie) waarin de symptomen (gedeeltelijk) verdwijnen. Ongeveer de helft van de mensen met RRMS ontwikkelen na ongeveer 10 jaar de secundair-progressieve vorm van MS (SPMS). Deze fase wordt gekenmerkt door geleidelijke en progressieve achteruitgang van neurologische symptomen met minimale terugvallen en herstelperiodes. In een klein deel van de mensen met MS, ongeveer 15%, wordt de ziekte gekarakteriseerd door progressieve verslechtering vanaf het beginstadium, ook wel bekend als primaire progressieve MS.

Pathologie van MS

Bij MS vinden er ontstekingen plaats in het brein en ruggenmerg wat als gevolg heeft dat de isolerende laag (myeline) om de zenuwbanen (axonen) wordt afgebroken, ook wel bekend als laesies of (sclerotische) plaques. Deze vettige laag om de zenuwbanen, de myeline, zorgt ervoor dat informatie (elektrische signalen, ook wel actiepotentialen genoemd) snel en efficiënt van neuron A naar neuron B kan komen, wat ervoor zorgt dat complexe taken kunnen worden uitgevoerd zoals het verwerken van informatie, praten en bewegen. Echter, de beschadigingen bij MS kunnen ervoor zorgen dat deze overdracht van informatie verstoord en of vertraagd wordt, waardoor er onder andere cognitieve en of bewegingsproblemen kunnen ontstaan. Ondanks dat myeline afbraak kan herstellen is dit vaak gedeeltelijk en kunnen de zenuwbanen en zelfs de zenuwcellen beschadigd raken en/of uiteindelijke verdwijnen. Ook zien we bij MS corticale laesies, aangezien ook in de hersenschors (cortex) myeline aanwezig is, en atrofie (krimpen) van het hersenweefsel.

Bij progressieve vormen van MS zien we een sneller beloop van deze neurodegeneratieve aspecten van MS, wat mogelijk verklaart waarom deze mensen sneller achteruit kunnen gaan. Vergeleken met 20 jaar geleden is het aantal opties voor medicatie flink gegroeid, met name voor RRMS, maar helaas is genezing en volledige remming van deze pathologische processen nog niet mogelijk.

Geavanceerde MRI-technieken om motorische stoornissen bij MS te onderzoeken
Beeldvorming met magnetische resonantie (MRI) is een belangrijk hulpmiddel voor de diagnose van MS maar ook voor de neurowetenschappen en heeft aanzienlijk bijgedragen aan het begrijpen van MS. Het klassieke pathologische kenmerk van MS, de inflammatoire en demyeliniserende witte stof laesies, kunnen worden gevisualiseerd met behulp van conventionele MR-sequenties in de dagelijkse neuro(radio)logische praktijk. Hoewel in de kliniek het visualiseren van focale witte stof laesies essentieel is voor de diagnose van MS en het monitoren van ziekteprogressie, blijft de associatie tussen laesies en klinische problemen beperkt. Waarbij bij de ene patiënt een grote hoeveelheid laesies kan leiden tot minimale motorproblemen, kunnen bij de ander enkele laesies al zorgen voor ernstige invaliditeit. Deze dissociatie tussen het aantal en volume van laesies en klinische presentatie is ook wel bekend als het klinisch-radiologisch paradox.

Met meer geavanceerde MRI-technieken is het mogelijk om beschadigingen in meer detail in kaart te brengen, zowel in de witte als de grijze stof. Waar enkel een matige associatie wordt gevonden tussen het laesie volume en de klinische achtergang, hangt atrofie (krimpen) van het hersenweefsel sterker samen met symptomen, ook al verklaart deze neurodegeneratie de klinische heterogeniteit nog steeds niet volledig. Omdat de hersenen een zeer complex systeem zijn met vele connecties kunnen symptomen wellicht beter begrepen worden door te bestuderen hoe bij MS de hersengebieden anders met elkaar interacteren en verbonden zijn, hetgeen wat met conventionele MRI-scans niet te zien is. Recente nieuwere beeldvormingstechnieken zoals diffusie gewogen MRI en functionele MRI hebben het mogelijk gemaakt om de structurele en functionele connectiviteit tussen verschillen hersengebieden te bestuderen en hebben ons veel nieuwe informatie over MS gegeven.

Met diffusie-gewogen MRI kunnen we meten hoe watermoleculen zich bewegen in het hersenweefsel. Hieruit kunnen we de micro structurele beschadigingen in laesies maar ook in hersenweefsel zonder laesies (normaal-ogende witte stof genoemd) afleiden. Met behulp van deze in onderzoek veel gebruikte methode kan worden bepaald in hoeverre de integriteit van belangrijke motor banen in de hersenen is aangedaan bij MS en dat deze schade relateert aan invaliditeit. Echter, met nieuwere diffusie methodes en MRI op zeer sterk magnetisch veld (7T) is het mogelijk om deze schade nog specifieker en sensitiever te detecteren, wat de mogelijkheid geeft om structurele veranderingen in een nog vroeger stadium van de ziekte te detecteren in mensen met minimale hersenschade en geen tot minimale invaliditeit.

Naast het in kaart brengen van de structurele schade in de hersenen en ruggenmerg is het is ook mogelijk om de hersenfunctie te onderzoeken met behulp van functionele MRI-technieken (fMRI). Met fMRI kan worden gemeten worden hoe actief een gebied is tijdens het uitvoeren van een bepaalde taak zoals het tikken van de vingers of bewegen van de voet, of tijdens rust. Vervolgens kan deze informatie gebruikt worden om te kijken welke gebieden tegelijkertijd actief zijn (functionele connectiviteit) en hoe deze anders met elkaar communiceren bij MS. Deze techniek en diffusie-gewogen MRI zijn zeer waardevol, aangezien de relatie tussen functionele en structurele verstoringen en motorische problemen helaas nog niet goed wordt begrepen, met name in een vroeg stadium van de ziekte.

Doelstelling van het proefschrift

Dit proefschrift had als doel nieuwe mechanismen te identificeren die gerelateerd zijn aan motorische stoornissen in vroege en latere fasen van MS. Daarom bestudeerden wij patronen van hersenweefsel schade en functionele veranderingen die ten grondslag zouden kunnen liggen aan problemen met het bewegen. Daarnaast is dit werk een van de eerste waar de armen en benen afzonderlijk werden bestudeerd bij MS.

Het proefschrift is onderverdeeld in twee voornaamste onderdelen, gebaseerd op twee studies in Amsterdam (hoofdstukken 2.1 en 2.2) en twee in Melbourne (hoofdstukken 3.1 en 3.2). Na een algemene introductie in hoofdstuk 1 hebben we in hoofdstuk 2.1 functionele veranderingen bestudeerd bij MS door te kijken naar de efficiëntie van het motorische netwerk in een groot cohort met mensen met milde en ernstige motorproblemen met 3T MRI. Dit onderzoek werd echter beperkt tot een algemene maat voor invaliditeit (de zogenaamde EDSS) en maar één meetmoment. Vervolgens hebben we daarom in hoofdstuk 2.2 gekeken naar de relatie van functionele hersenveranderingen en aparte metingen van het functioneren van handen en benen, en werd ook over een periode van vijf jaar gemeten. In hoofdstuk 3 hebben we gekeken naar een tweede cohort wat bestond uit mensen met geen tot minimale klinische invaliditeit en vroege MS. Hier hebben we gebruik gemaakt van 7T MRI en nog sensitievere hand en looptesten, wat ons de mogelijkheid gaf om subtiele motorproblemen en functionele (hoofdstuk 3.1) en structurele (hoofdstuk 3.2) veranderingen in de hersenen te onderzoeken in deze groep mensen met zeer weinig schade en symptomen.

Hoofdstuk 2.1

Het doel van onze eerste studie was om de functie van het motorische hersennetwerk te bestuderen in mensen met MS, waarin we mensen met milde en meer ernstigere motorproblemen vergeleken met gezonde vrijwilligers. Hiervoor hebben we data gebruikt van het Amsterdam MS cohort. Mensen werden gevraagd mee te doen aan meerdere klinische testen waaronder neurologisch onderzoek (de EDSS, een schaal van invaliditeit vaak gebruikt in de kliniek), en een MRI-onderzoek. Tijdens de MRI-sessie hebben we verschillende soorten scans verzameld, waaronder een scan waarbij we de hersenfunctie

tijdens rust hebben gemeten. Deze scan hebben we gebruikt om te kijken naar de functionele connecties tussen verschillende motor gebieden. Om de efficiëntie van het motor systeem in kaart te brengen hebben we gebruik gemaakt van geavanceerdere netwerkanalyses, de zogenaamde "graaf analyse". De functionele connectiviteit vormde de basis voor deze geavanceerdere netwerkanalyses van het motor systeem.

Wij vonden dat deze efficiëntie omhooggaat bij mensen met MS, maar dan alleen bij die mensen met ernstigere bewegingsstoornissen (die niet kunnen lopen zonder assistentie of een hulpmiddel) in vergelijking met mensen met minder ernstige motorproblemen (kunnen lopen zonder hulp) en gezonde vrijwilligers. Mensen met milde bewegingsproblemen hadden een normaal motorisch hersennetwerk, oftewel de functie van dit netwerk was vergelijkbaar met een gezond brein. We hebben ook verder ingezoomd om te kijken welke gebied met name verantwoordelijk was voor dit effect, en dat was de primaire somatosensorische cortex (S1). Dit gebied is verantwoordelijk voor het verwerken van sensorische informatie afkomstig uit het lichaam dat via het ruggenmerg naar de hersenen wordt doorgestuurd en het doorzenden van informatie naar andere hersengebieden. Dit gebied vertoonde sterkere connecties met andere motor gebieden in het netwerk wat gerelateerd was aan de ernst van de motorische afwijkingen. Mogelijk is deze sterke connectiviteit een indicatie ("marker") voor een verstoorde communicatie binnen de hersenen, waardoor het hele netwerk verstoord wordt, wat gerelateerd is aan slechter lopen. Dit zou bijvoorbeeld vergeleken kunnen worden met ruis in het signaal van een radiozender, waardoor de informatie in het signaal niet goed overgebracht kan worden. Deze interpretatie blijft echter speculatief en zal in vervolgonderzoek moeten worden bewezen.

Hoofdstuk 2.2

Het doel van de volgende stap van het onderzoek was deze netwerkveranderingen te volgen over tijd en te relateren aan klinische progressie (het verslechteren van bewegen), wat apart werd gemeten in de armen en benen. Om het beloop van de ziekte te kunnen onderzoeken kwamen deelnemers van het Amsterdam MS cohort terug na 5 jaar en ondergingen nogmaals de neurologische onderzoeken en MRI-scans. Naast de veranderingen in de netwerk topologie waren we ook geïnteresseerd in de veranderingen in de netwerk dynamiek. Deze "dynamiek" is een nieuwe techniek om hersenfunctie nog gedetailleerder te bekijken. Hersengebieden zijn namelijk niet constant met elkaar verbonden, maar in werkelijkheid fluctueert de sterkte van verbondenheid/communicatie over tijd. Er zijn dus periodes van sterke en zwakke verbindingen, wat ook nodig is om de hersenfunctie te kunnen switchen tussen netwerken en om verschillende taken te kunnen uitvoeren.

Deze dynamische patronen hebben wij in dit onderzoek voor het eerst gerelateerd aan motorische problemen bij MS. We vonden onder andere dat een verhoogde dynamiek van het motorisch netwerk te zien was bij mensen die over 5 jaar tijd klinische achteruitgang lieten zien. Deze verhoging duidt op een instabieler netwerk, wat dus voorspellend lijkt te zijn voor klinische progressie. Verschillende gebieden waren verantwoordelijk voor de

functionele achteruitgang van de armen en benen. Zo vonden we dat de supplementaire motorische schors (SMA) een belangrijke rol speelde bij de achteruitgang van loop functie. Dit gebied is normaliter betrokken bij het plannen van bewegen, en is bij complexere processen betrokken dan primaire gebieden zoals bijvoorbeeld S1. Daarnaast bleek een verstoorde dynamiek van de thalamus met name een rol te spelen in de achteruitgang van armfunctie. De thalamus is een van de belangrijkste "schakelstations" of "hubs" in ons brein, vergelijkbaar met Schiphol in het Europees vliegverkeer en heeft daarom een grote invloed op het functioneren van het hele netwerk. Deze resultaten wijzen erop dat de mechanismen onderliggend aan achteruitgang van arm en beenfunctie mogelijk anders zijn, wat ook zou kunnen verklaren waarom deze meestal niet tegelijk optreden bij MS. Daarnaast lijkt het dus dat een hogere dynamiek een onstabiel netwerk zou kunnen reflecteren, wat er vervolgens voor kan zorgen dat het netwerk gevoeliger is voor verdere verstoring, wat zou kunnen leiden tot klinische achteruitgang.

Hoofdstuk 3.1

In hoofdstuk 2 hebben wij verstoringen in het functionele motor netwerk gemeten bij mensen met MS met vrij ernstige motorproblemen. Vervolgens wilden wij weten welke veranderingen in de hersenen zichtbaar zijn bij mensen met MS met zeer milde bewegingsproblemen en een vroege ziekteduur. Daarom hebben wij voor hoofdstuk 3 gebruik gemaakt van sensitieve klinische metingen van loop en arm functie om subtiele veranderingen te kunnen detecteren en een 7T MRI-scanner om zeer gedetailleerde hoge resolutie beeldvorming van de hersenen te kunnen verkrijgen. In hoofdstuk 3.1 waren we vooral geïnteresseerd in hersenfunctie, en dan vooral in de patronen van hersenactivatie tijdens het uitvoeren van een complexe motor taak met de hand en de voet. Men werd gevraagd om een harmonische bewegende lijn te volgen door zachtjes te knijpen met de vingers en weer los te laten of door hun enkel voorzichtig heen en weer te bewegen. Om het bewegend doel nauwkeurig te volgen was dus de integratie van proprioceptieve en visuele input erg belangrijk.

Mensen met MS waren slechter in het uitvoeren van de enkel/voet taak dan gezonde vrijwilligers. Tijdens het uitvoeren van deze taak vertoonden mensen met MS minder activiteit ("lagere activatie") dan gezonde vrijwilligers in verschillende gebieden in het brein betrokken bij visuomotorische integratie, oftewel gebieden betrokken bij het combineren van visuele en motorische informatie. Lagere activatie tijdens het bewegen correleerde met een slechtere prestatie van het uitvoeren van deze motor taak en meer structurele schade in het brein. Ondanks dat mensen met MS niet slechter presteerden tijdens de hand taak, vonden we alsnog wel een lagere activatie in de visuele cortex. Deze verandering kunnen een vroege indicatie zijn van een verstoorde hersenfunctie, en dat armfunctie pas later achteruit zou kunnen gaan. Deze latere achteruitgang van armfunctie is ook wat er klinisch vaker is gerapporteerd in eerder onderzoek. Potentieel liggen er dus verschillende functionele mechanismen ten grondslag aan problemen met de benen en armen. Met behulp van ons crosssectioneel onderzoek hebben we kunnen aantonen dat stoornissen

in motor controle bij mensen in MS relateert aan verstoringen in visuomotorische integratie in de hersenen. Ook hebben we aangetoond dat met een hoger magnetisch veld (7T MRI) en een complexe motor taak zeer subtiele veranderingen in beweging en hersenactiviteit gedetecteerd kunnen worden bij mensen met MS met minimale invaliditeit. Vervolgonderzoek is nu nodig om deze bevindingen te valideren in longitudinaal onderzoek, om te bekijken of deze veranderingen voorspellend zijn voor latere achteruitgang.

Hoofdstuk 3.2

In hoofdstuk 3.2 hebben wij hetzelfde cohort bekeken als in hoofdstuk 3.1, maar waren we hier met name geïnteresseerd in het zo nauwkeurig mogelijk in kaart brengen van schade in belangrijke motorpaden in de hersenen. We wilden deze patronen van schade relateren aan subtiele verandering in handfunctie en lopen. Om schade te onderzoeken hebben we gebruik gemaakt van geavanceerde diffusie maten geassocieerd met de dichtheid van vezelbundels. Deze techniek kan mogelijk een gevoelige indicatie geven van het verlies en/of krimpen van axonen (zenuwbanen). Onze resultaten lieten zeer uitgebreide veranderingen zien in de hersenen van deze mensen met milde MS. Ondanks de minimale beperkingen in motorfunctie en de minimale aanwezigheid van laesies vonden we een uitgebreid patroon van schade. Deze schade bestond uit een kleinere cross-sectie van de vezelbundels (oftewel, het krimpen van zenuwbanen) in drie belangrijke motorbanen in de hersenen: de verbindingen van de motor cortex naar het ruggenmerg, de connecties tussen de hemisferen en de verbindingen tussen het cerebellum en de thalamus. De ernst van de axonale schade relateerde aan meer foutieve handbewegingen tijdens de motor taak en een veranderd looppatroon (kleinere stapbreedte, korter contact van de voet op de grond en langere stand op twee benen). Toekomstig onderzoek moet uitwijzen of deze technieken mogelijk in de kliniek gebruikt kunnen worden in een dergelijk vroeg stadium van MS.

Samenvatting

In dit proefschrift hebben wij de mechanismen die gerelateerd zijn aan motorische stoornissen in vroege en latere fasen van MS onderzocht met behulp van geavanceerde MRI-technieken en verschillende klinische maten van arm en been functie. Ons onderzoek analyseerde functionele MRI-scans tijdens rust (hoofdstukken 2.1 en 2.2) en taak (hoofdstuk 3.1), evenals diffusie gewogen beeldvorming (hoofdstuk 3.2) en 7T MRI (hoofdstukken 3.1 en 3.2). Onze bevindingen lieten een duidelijk verband zien tussen de ontwikkeling van motorische stoornissen en veranderingen van de structuur en functie in het motor systeem van de hersenen bij MS. Door het motor systeem in zijn geheel te bestuderen met behulp van netwerkanalyses in een groot cohort van mensen met MS, identificeerden we functionele stoornissen die alleen aanwezig waren in mensen met MS met ernstige problemen met lopen (hoofdstuk 2.1) en vonden we dat stoornissen in de netwerk dynamiek voorspellend waren voor de achteruitgang van arm en been functie (hoofdstuk 2.2). Verder vonden we met behulp van 7T MRI en sensitieve metingen arm en loop functie in een cohort van patiënten met minimale motorische beperkingen een verband tussen veranderingen in de functie (hoofdstuk 3.1) en microstructuur (hoofdstuk 3.2) van het motorische systeem en de aanwezigheid van subtiele stoornissen in handfunctie en lopen.

De belangrijkste bevindingen van dit proefschrift zijn:

1. Functionele stoornissen in het motorische hersennetwerk bij mensen met MS met ernstigere invaliditeit

- Van de 222 mensen met MS vertoonde 185 minimale invaliditeit en 37 ernstigere problemen met lopen.
- Mensen met geen tot milde bewegingsproblemen (kunnen lopen zonder hulp) vertoonden een normaal motorisch netwerk vergeleken met gezonde vrijwilligers.
- Mensen met ernstigere bewegingsproblemen (niet kunnen lopen zonder assistentie of hulpmiddel) vertoonden verhoogde netwerk efficiëntie en connectiviteit.
- De primaire somatosensorische cortex (S1), een hersengebied wat belangrijk is voor het ontvangen en integreren van sensorische informatie, speelde een belangrijke rol bij het verstoorde motorische netwerk bij mensen met ernstigere bewegingsproblemen.
- Van de 214 mensen met MS vertoonden 24 achteruitgang van de bovenste ledematen en 67 achteruitgang van de onderste ledematen over een periode van vijf jaar.
- Achteruitgang kon worden voorspeld door een verhoogde netwerk dynamiek.
- De thalamus en SMA speelden een belangrijke rol in deze verstoorde stabiliteit van het motorische netwerk.

2. Veranderingen in functionele hersenactivatie bij mensen met vroege MS met milde invaliditeit

- Mensen met vroege MS vertoonden vertraagde en foutieve bewegingen van de benen maar geen verandering in handfunctie.
- Lagere hersenactivatie werd gezien tijdens het bewegen van de voet in het cerebellum, visuele cortex en pariëtale cortex, gebieden belangrijk voor visuomotorische integratie.
- Lagere activatie correleerde met een slechtere prestatie van het uitvoeren van de motor taak en meer structurele schade in het brein.
- Tijdens het (normaal) bewegen van de handen werd een verlaagde activiteit in de visuele cortex gezien.

3. Hersenschade bij mensen met MS met milde invaliditeit

- Mensen met vroege MS met milde motorproblemen vertoonden zeer uitgebreide schade in belangrijke motorbanen in de hersenen.
- Dit patroon van schade was uitgebreider dan de focale laesies.
- De ernst van schade was geassocieerd met een slechtere handfunctie en veranderd looppatroon.